**Pharmacokinetics-Pharmacodynamics of Antifungal Agents in the Central Nervous System**

KE Stott1, 2, W Hope1

1 Antimicrobial Pharmacodynamics and Therapeutics Laboratory, Department of Molecular and Clinical Pharmacology, University of Liverpool, UK

2 Malawi-Liverpool-Wellcome Trust Clinical Research Programme, Blantyre, Malawi

**Abstract**

**Introduction**

Mortality from invasive fungal disease involving the central nervous system (CNS) is excessive. Achieving therapeutic drug concentrations at the site of infection within the CNS is always difficult and its evaluation is complex due to anatomical barriers and variable pathophysiological lesions.

**Areas covered**

This review provides an updated summary of the CNS PK of antifungal therapies. It considers factors that influence the success of antifungal regimens for CNS infection as well as preclinical and clinical data that quantify antifungal pharmacokinetics (PK) in the CNS. Furthermore, it presents state-of-the-art technologies to enhance the clinical use of existing antifungal drugs, and introduces novel antifungal drugs in development.

**Expert opinion**

The antifungal drugs currently available are either suboptimal, or are being used suboptimally, for CNS disease. Therapeutic drug monitoring is mandatory to enhance their effectiveness. Novel drugs in development may offer more efficacious options. In all cases, contemporary technologies to assess CNS PK offer the opportunity to enhance our understanding and use of antifungal drugs for CNS fungal disease.

**Keywords:**

Antifungal, central nervous system, cerebrospinal fluid, brain, pharmacokinetics, distribution.

**Introduction**

Mortality from invasive fungal diseases involving the central nervous system (CNS) frequently exceeds 50% (1). There are unique challenges for drug penetration within the CNS. The blood-brain barrier and blood-cerebrospinal fluid barrier create an obstacle for free diffusion of compounds into the CNS. Hence, plasma drug concentrations are not reliable surrogates for concentrations within diseased areas within the brain (2). Complex pathophysiology and nonspecific clinical presentations often lead to a late diagnosis of CNS mycoses and this further compromises pharmacological treatment. This review provides a summary of current knowledge of the CNS pharmacokinetics (PK) of antifungal therapies.

**1. CNS PK of antifungal drugs**

The CNS is comprised of multiple sub-compartments that include the cerebrospinal fluid (CSF), cerebral parenchyma, ventricles and meninges (3). These areas are pharmacologically distinct. A number issues are pertinent for a complete understanding of the potential utility of an antifungal agent for the treatment of CNS mycoses. First, pathological changes within the CNS resulting from fungal invasion; second, physicochemical drug properties that influence the extent of partitioning into the CNS and areas of diseased tissue; and finally, PK variability.

**1.1 Pathophysiology of CNS fungal infections**

Fungi vary considerably in the extent to which they are neurotropic. For some pathogens, such as *Cryptococcus neoformans* and *Cladophiolophra*, involvement of the CNS is so characteristic that it must be actively excluded if a diagnosis is established from a non-CNS site. For others, such as *Aspergillus* spp., CNS involvement is well characterised but is not invariably present. In addition, involvement of the CNS may depend on the host. For example, hematogenous *Candida* meningoencephalitis (HCME) is a disease that is characteristic of premature neonates, but highly unusual in adults. Table 1 summarises the considerable pathophysiological differences between CNS mycoses.

The nature and extent of underlying immunological deficits also has an important impact on patterns of infection and the cadence of clinical disease. For example, profoundly immunosuppressed patients with invasive CNS aspergillosis often present with a stroke-like illness resulting from cerebral infarction. This results from invasion and thrombosis of cerebral arteries by *Aspergillus* spp. In contrast, patients with chronic sinus aspergillosis may develop CNS disease via direct hyphal invasion through the lamina papyracea. These patients often only have mild immunological deficits (e.g. diabetes, low dose corticosteroid treatment) with clinical signs and symptoms that develop over many months. The histopathology of these two diseases is distinct. Cerebrovascular aspergillosis is associated with a paucity of inflammation, infarction and cerebral haemorrhage. In contrast, chronic *Aspergillus* sinusitis is associated with florid pyogranulomatous inflammation.

Systemic physiological changes associated with CNS mycoses may also have an impact on the PK of antifungal drugs. Systemic infection alters cerebral blood flow, blood and tissue pH and intra- and extra-cellular fluid volumes, all of which exert profound influence on systemic PK and drug penetration into the CNS (4-6). Meningeal inflammation may increase the concentration of antimicrobial agents in the CSF by over 10-fold (7). In some cases the blood-brain barrier (BBB) is completely disrupted, allowing unfettered access of the drug to the pathogen.

**1.2 Physicochemical properties of the drug**

The CNS is protected from many circulating xenobiotics by the presence of blood tissue barriers that limit diffusion from the endovascular compartment into the CSF and brain parenchyma. A summary of antifungal drug characteristics that influence the degree to which drugs partition across the BBB and the blood-cerebrospinal fluid barrier (BCSFB) is presented in Table 2. Tight cellular connections in the BBB and BCSFB (approximately 2 nm) prevent the passive diffusion of large compounds into the CNS (3). Efficient diffusion is possible only to an upper molecular weight limit of 300–400 g/mol (8).

The lipophilicity of compounds influences the extent of CNS partitioning (4, 9). Drug lipophilicity is quantified as the partition coefficient (LogP) between aqueous and lipophilic phases (usually determined with water and octanol). A more physiologically relevant expression of lipophilicity is LogD, which accounts for the fact that many drugs are ionised at physiological pH. Compounds that are not ionized at physiological pH have the greatest lipophilicity and better penetrate the BBB and BCSFB (4, 10). LogP and LogD values of approximately 2–4 are associated with optimal BBB penetration (10, 11). Protein binding also influences the degree of partitioning into the CNS. Passive diffusion of molecules into the CNS depends on a concentration gradient between unbound drug in the plasma and that in the brain (12). Protein binding is often inversely correlated with LogP.

Finally, efflux pumps in the BBB and BCSFB may remove compounds from the CNS via an energy-dependent process (3, 4). P-glycoprotein is a membrane-bound efflux pump with an affinity for lipophilic molecules. Some triazole agents (itraconazole, posaconazole and isavuconazole (13)) are substrates for P glycoprotein, while other triazoles (fluconazole and voriconazole) are not. There is conflicting evidence regarding the role of P-glycoprotein in the efflux of amphotericin B from the CNS (14, 15).

**1.3 PK variability**

Several antifungal drugs exhibit non-linear and/or highly variable PK. The PK of itraconazole, voriconazole, posaconazole and flucytosine are sufficiently variable to warrant routine therapeutic drug monitoring in the context of invasive fungal disease, regardless of whether there is CNS involvement (16). This variability is inevitably more extreme when penetration into the CNS is considered, since it is amplified by barriers such as the BBB and BCSFB as well as pathological changes within the CNS itself. Thus, while plasma drug concentrations are highly variable, CNS drug concentrations likely more so, and the former are an unreliable surrogate for the latter.

The prediction and evaluation of the CNS PK of antifungals is thereby complex. Inter- and intra-individual variability in each contributing factor extends this complexity (4, 16). Ideally, the information required to establish robust pharmacokinetic-pharmacodynamic (PK-PD) targets at the site of infection in the CNS includes all of the points listed in Box 1. Using this information, data can be modelled using a variety of approaches to predict human CNS PK. These approaches can broadly be categorized into classical population PK modelling (the ‘top-down’ approach) and physiologically based PK (PBPK) modelling (the ‘bottom-up’ approach) (17). For example, population PK models have been constructed from preclinical investigations of micafungin (18) and anidulafungin (19), with Monte Carlo simulation used to bridge the results to neonatal populations at risk of *Candida* meningoencephalitis. Whilst PBPK models describing the CNS PK of antifungals are scarce, examples of the application of PK modelling to the prediction of CNS PK exist in the general pharmacology literature. A multi-compartmental PBPK model has been shown to adequately describe the PK profiles of nine structurally diverse drugs in the plasma, brain extracellular fluid and CSF of rats (20). The model was additionally able to predict human concentration-time profiles in brain compartments (20, 21). PBPK models can also predict the influence of genetic polymorphisms on CNS PK and PD (22).

**2. Evaluation of data relating to the CNS PK of antifungal drugs**

Studies of the CNS penetration of antifungal drugs in humans are often limited. Firstly, drug concentrations in CSF, which is the only readily available biological matrix in clinical studies, may not be representative of concentrations in other CNS sub-compartments. Secondly, studies tend to report point estimates of CNS drug partitioning rather than the trend of drug concentrations over time (i.e. area under the concentration-time curve; AUC) in the plasma and the CNS.

For obvious ethical and technical reasons, quantification of drug in the CNS using conventional methods such as collecting samples of tissue for liquid chromatography-mass spectrometry (LC-MS) or bioassay in clinical studies is challenging (see, for example, (23, 24)). Achieving this at several time points throughout the dosing interval is even more so (2). Information of this type is invariably derived from preclinical models. Using more contemporary technologies, clinically relevant estimation of CNS drug partitioning is becoming easier. Examples of these technologies are intracerebral microdialysis (12) and non-invasive imaging techniques such as positron emission tomography (PET) (25) and magnetic resonance (MR) imaging (26). Key findings that preclinical and clinical studies have provided are presented in the following section of this review, alongside a review of their clinical application.

1. **The triazoles**

Fluconazole

Fluconazole has physicochemical properties that enable it to traverse the BBB and BCSFB (Table 2). The equilibration of fluconazole between plasma and cerebral extracellular fluid occurs rapidly and is independent of dose (27). Relatively high concentrations are found in the CSF. PK studies in rabbits and adult Rhesus monkeys demonstrated mean CSF:plasma AUC ratios of 0.84 and 0.86 respectively, with a long half-life in the CSF of approximately 27 hours (28, 29).

Early human CNS PK studies using traditional bioassay or LC-MS reported a range of partition ratios from 0.52 – 0.89 (23, 30). Subsequently, a combined approach using positron emission tomography (PET) scanning and plasma PK sampling demonstrated an approximately uniform distribution of fluconazole in the healthy human brain, with a brain:plasma penetration ratio of 1.31 (25). This was corroborated by an analysis of 4 patients undergoing brain tumour excision in whom healthy brain parenchyma was also removed. In these patients, HPLC demonstrated a mean healthy brain:plasma fluconazole ratio of 1.33 (24).

A fluconazole regimen of 1200mg/day for the 2 week induction phase of treatment for cryptococcal meningoencephalitis is recommended if polyenes are unavailable (31). This dosage is more rapidly fungicidal than 800mg/day (32). The addition of flucytosine to fluconazole is recommended because it reduces mortality (33). These oral regimens for cryptococcal meningitis are pragmatic recommendations given the unavailability of polyenes in many regions with a high burden of cryptococcal meningitis, despite broad agreement that amphotericin B deoxycholate is currently the agent of choice (34, 35). Fluconazole is also used at dosages of 400-1200mg/day to treat CNS infection with *Coccidioides* (36, 37).

Itraconazole

Itraconazole concentrations in CSF are negligible, with CSF:plasma concentration ratios of <0.002 to 0.12 in preclinical models (2, 38-40). Even in the setting of infection and inflammation, itraconazole is undetectable in rabbit CSF (40). Low CSF concentrations have been attributed to rapid binding to red blood cells and circulating plasma proteins, inhibiting BCSFB penetration (41). However, murine experiments have demonstrated rapid, dose-dependent penetration and linear accumulation of itraconazole in brain tissue up to 8 minutes post-dose, implying that it does cross the BBB (42). In a murine model of CNS histoplasmosis, itraconazole levels were almost universally undetectable in brain tissue 3 hours post-dose (43). Itraconazole has a strong affinity for P-glycoprotein, which results in rapid efflux from the CNS such that half-life in cerebral tissue may be considerably shorter than that in plasma (0.4h versus 5h, respectively) (42, 44).

The efficacy of itraconazole for the treatment of cryptococcal meningoencephalitis (40), CNS aspergillosis (45) and *Coccidioides* meningitis (38) is well established in laboratory animal models. The apparent discrepancy between CNS drug concentrations and therapeutic efficacy of itraconazole in CNS fungal infections may be due to a combination of higher drug concentrations in the brain tissue than in CSF, and the relatively low minimum inhibitory concentration of itraconazole against target fungi such as *Candida* (46) and *Histoplasma* (43). In addition, the pharmacologically active metabolite of itraconazole, hydroxyl-itraconazole, has been detected in brain parenchyma with greater consistency than has the parent drug (43). In humans, itraconazole is effective for primary prophylaxis of cryptococcal meningitis, with a relative risk of incident cryptococcal meningitis of 0.12 (95% confidence interval, 0.03, 0.51) versus placebo (47). Inconsistent success rates have been reported for the treatment of cryptococcal meningitis with itraconazole. The treatment of 6 patients with 200mg/day resulted in therapeutic failure (deterioration or death) in 3 (50%) (48). Of 20 evaluable patients with culture- or antigen-diagnosed cryptococcal meningitis treated with itraconazole 400mg/day, 13 (65%) achieved clinical and microbiological cure (49). Cryptococcal meningitis progressed in 2 patients receiving itraconazole 600mg/day (50). After recovery from cryptococcal meningitis, maintenance therapy with itraconazole is associated with significantly more relapses of cryptococcal meningitis compared with fluconazole (51).

Itraconazole is not included in current cryptococcal meningitis treatment guidelines from the World Health Organisation (31), though it may be used as prophylaxis against cryptococcal meningitis, particularly in patients with CD4 counts <100 cells/µL (52). Despite the availability of newer antifungal agents, the oral bioavailability and broad spectrum of antifungal activity of itraconazole mean that it remains a useful drug for the management of invasive mycoses worldwide (53). It is recommended as second-line therapy for CNS infection with *Candida*, *Aspergillus, Histoplasma* and *Coccidioides* spp. (36).

Voriconazole

 Voriconazole has a relatively low molecular weight (349 g/mol), is moderately lipophilic and is a weak substrate for P-gp (Table 1). It exhibits good CNS penetration. In guinea pigs, 10 mg/kg voriconazole penetrates the BBB into brain parenchyma rapidly, reaching peak concentrations of 6.8 µg/g and 2.7 µg/g 15 minutes and 1 hour post dose, respectively (54). The CSF:plasma ratio in healthy animals is 0.68 (54).

Studies in humans have demonstrated that voriconazole penetrates the BBB and BCSFB, including into brain parenchyma (55) and cerebral abscesses (56). A wide range of concentrations in human CSF range have been reported, from 0.08 to 3.93 micrograms/mL in the initial 10 h post voriconazole administration. CSF:plasma ratios are 0.22-1.0 (54). This wide range is likely due to the extensive PK variability of voriconazole (16, 57-61). In contrast to CSF, concentrations in the brain are ≥2-fold plasma levels (26). A study that used fluorine-19 MR spectroscopy reported steady-state brain:plasma voriconazole concentration ratios of 3.0 (90% CI 1.9 – 4.7) pre-dose and 1.9 (90% CI 1.2 – 3.0) post-dose in healthy adult men (26).

Voriconazole exhibits potent activity against *Aspergillus* spp. (Table 2) and is a standard-of-care for CNS aspergillosis (62). It is also indicated for CNS infection with *Scedosporium apiospermum* complex and fluconazole-resistant *Candida* spp. (Table 2), the latter being primarily confined to the neonatal setting.

Posaconazole

 Posaconazole is structurally similar to itraconazole, but less lipophilic (2). It is both a substrate and an inhibitor of P-gp *in vitro* (63), although polymorphisms of P-gp do not affect posaconazole AUC *in vivo* (64). The oral suspension of posaconazole exhibits variable bioavailability and inconsistent PK (65, 66). Alternative tablet, capsule and intravenous formulations have been partly successful in addressing these issues (67-71). Based on the physicochemical properties of posaconazole, poor CNS penetration is expected (Table 2). In murine models of infection with *C. gattii* and *Fonseca monophora*, mean posaconazole brain:serum concentration ratios are < 0.54 with dosages of 10-20 mg/kg. These increase to 0.69-0.84 following dosages of 40 mg/kg (72, 73).

Human CNS PK data for posaconazole is sparse and limited to case reports. The CSF:plasma concentration ratios range from 0 (single CSF sample analysed in each case) (74, 75) to <0.009 (in 6 serial CSF samples) (76) in patients without significant meningeal inflammation. In patients with bacterial meningitis and cerebral fungal infection, CSF:plasma posaconazole concentration ratios as high as 0.44 and 2.37 are reported, suggesting that meningeal inflammation may improve CNS penetration (75). Posaconazole penetrates into fungal brain abscesses (75).

Posaconazole is not recommended as a first-line agent for any fungal infection of the CNS. Posaconazole is indicated as second line therapy for coccidioidomycosis (77), and mucormycosis (78) and as salvage therapy for invasive aspergillosis (79, 80) and histoplasmosis (81) (Table 2, (36)).

Isavuconazole

 Isavuconazole is structurally similar to fluconazole and voriconazole, although it has a higher molecular weight and LogD value (Table 2). Studies in healthy rats suggest that the mean brain:plasma isavuconazole concentration ratio is 1.8 (82). Following administration of 14C-labeled isavuconazonium sulphate, radioactivity in brain tissue increases in proportion with radioactivity in blood, and reaches a maximum at 2h post-dose before declining to undetectable levels 24h post-dose (82). Similar CNS penetration is evident in a murine model of cryptococcal meningitis with brain:plasma AUC ratios of approximately 1.35 (83).

 Data describing the PK of isavuconazole in the human CNS are scant. However, of 27 patients with CNS fungal infection with aspergillosis, cryptococcosis or mucorales treated with isavuconazole, survival at 6 months was 50% (84).

**3. Polyenes**

There are several formulations of amphotericin B (AmB) available for clinical use: amphotericin B deoxycholate (DAmB), liposomal amphotericin B (LAmB), amphotericin B lipid complex (ABLC) and amphotericin B colloidal dispersion (ABCD) (36, 85). ABCD is no longer marketed in many countries. All approved formulations are administered intravenously. The large molecular weight of AmB is likely to be the primary reason for its relatively poor CNS penetration (86, 87) (Table 2). An *in vitro* model of the BBB demonstrated that permeability to both LAmB and DAmB was significantly increased in response to enlargement of endothelial cell junctions via exposure to either tumour necrosis factor alpha or lipopolysaccharide (88). In addition, AmB may be a substrate for efflux pumps at the BBB (14, 15).

In a rabbit model of CNS *C. albicans* infection, CSF:plasma ratios ≤ 0.02 and brain:plasma ratios ≤ 0.27 were reported for all four AmB formulations (86). The CNS penetration ratios of all formulations are higher in the setting of CNS infection compared with healthy brain tissue (maximum AmB CSF:plasma ratio 0.03 with ABCD and ABLC; maximum brain:plasma ratio 0.11 with ABCD) (86).

LAmB has the lowest CNS penetration ratio of all formulations despite its smaller particle size (Table 2). However, the PK of LAmB is characterised by serum concentrations 30-fold greater than other AmB formulations. Thus, the absolute concentration of AmB in brain tissue following LAmB administration is ultimately several times higher than following administration of the other formulations (3, 86). An immunohistochemistry study of LAmB in brain sections of mice with cryptococcal meningitis revealed that amphotericin B was present in both intravascular and perivascular spaces (89). The proportion of liposome-associated AmB versus free drug in brain tissue after administration of LAmB is unknown.

After as few as 3 dosages of DAmB, there is prolonged mean residence time of AmB in rabbit brain tissue despite negligible concentrations in CSF (90). In human studies of LAmB PK, CSF:plasma concentration ratios of 0.001 have been reported (91). Autopsy studies have found concentrations of AmB in human cerebral cortex to be lower than those in liver, spleen, kidney and lung, with no significant difference between the concentrations detected after administration of LAmB and ABCD (92).

Despite consistent reports of low or undetectable CNS concentrations of AmB, its efficacy against many CNS mycoses in humans, including cryptococcal meningitis, is well established (93-96). A likely explanation for this is that amphotericin B concentrations are high in the meninges where the yeast predominantly resides. Hence there is co-localisation of drug and pathogen, although to date this has been difficult to verify.

The clinical utility of polyenes in regions of the world with high burden of fungal CNS infection, in particular cryptococcal meningitis but also CNS histoplasmosis, murcomycosis, sporotrichosis and trichosporonosis, is limited by the requirement for intravenous administration, prolonged inpatient admission and close monitoring for toxicity.

**4. Flucytosine**

 Flucytosine has several attributes that are conducive to high CNS penetration including low molecular weight, low protein binding and polarity (Table 2). High dose-proportional concentrations are consistently recorded in both brain parenchyma (97) and CSF, with CSF:plasma ratios 0.74 - 0.84 in humans from 1-2 hours after dosing (98, 99). The activity of flucytosine appears to be time- rather than concentration-dependent (98, 100, 101).

Flucytosine is active against *Cryptococcus* spp., most *Candida* species and has some activity against *Aspergillus* species (16, 31). Its use in monotherapy is widely thought to be precluded by the development of resistance, which has been consistently documented *in vitro* (102-104). While reports on the clinical use of flucytosine monotherapy are sparse, a case series of 23 patients with cryptococcal meningitis treated with flucytosine monotherapy reported that of 12 patients who failed therapy, the development of resistance was implicated in 50% (105).

In practice, flucytosine is used in combination therapy. In this setting, flucytosine is vital to secure optimal outcomes in cryptococcal meningitis: Trials comparing DAmB alone with DAmB in combination with either flucytosine or fluconazole have demonstrated superior outcomes in the flucytosine combination arms, in terms of CSF sterilisation (106, 107) and mortality (106). Its addition to AmB facilitates reduced treatment durations for cryptococcal meningitis (108). 5FC may also have a role in HCME, where its addition to AmB is at least additive and may be synergistic in preclinical models (109, 110). A retrospective study of humans with *Candida* meningitis reported survival in 15 of 17 patients treated with the combination of flucytosine and AmB (111). This combination is recommended for *Candida* meningitis in children (112) though caution is advised because of the risk of toxicity, particularly in premature neonates (113).

**5. Echinocandins**

 Caspofungin, micafungin and anidulafungin do not extensively partition into the CNS due to their large molecular weight and high level of protein binding (Table 2, (114)). Micafungin achieves brain:plasma concentration ratios <0.01 at doses of 0.5 – 2mg/kg in healthy rabbits (115). In rabbits with HCME, micafungin penetrates all CNS compartments (cerebrum, cerebellum, spinal cord, meninges and CSF) in a dose-dependent fashion, but only with dosages >2mg/kg (18). The presence of CNS infection and inflammation does not increase the CNS penetration of micafungin (18). Brain tissue concentrations of anidulafungin also increase in a dose-dependent manner, achieving brain:plasma ratios of 0.10-0.12 at doses of 0.5-10 mg/kg in neutropenic rabbits with disseminated *C. albicans* infection (116). Studies of radiolabelled caspofungin at dosages of 1-2mg/kg demonstrate brain:plasma penetration ratios of <0.09 in rodents (117). For all three echinocandins, penetration into each of lung, liver, spleen and kidney exceeds that into brain tissue (115-117).

Human CNS PK data for echinocandins is limited to case reports. A patient with a cerebral mass treated with micafungin at a dosage of 100mg q24h achieved a brain:plasma concentration ratio of 0.18, 23 hours after dosing (118). A patient dosed with micafungin 300 mg/day for CNS aspergillosis achieved a CSF:plasma ratio of 0.0005 (119). A case report of a patient treated for *Candida* endocarditis describes the development of new cerebral abscesses during caspofungin treatment (120).

There is experimental evidence from well characterised rabbit models of HCME that anidulafungin and micafungin are potentially effective agents (18, 19). The weight-based dosages predicted for efficacy from PK-PD bridging studies are in excess of those recommended in adults for invasive candidiasis. This finding promoted further clinical studies of micafungin that included dosages up to 15 mg/kg. The European Society of Clinical Microbiology and Infectious Diseases recommends that a dosage of 10mg/kg is considered for neonatal HCME (18). Echinocandins may also be considered as an adjunct to other first-line antifungal agents for the treatment of CNS infection caused by *Aspergillus* spp., *Scedosporium apiospermum* complexand *Lemontospora* *prolificans*. (Table 1).

**6. Expert opinion**

The antifungal drugs currently available are either suboptimal, or are being used suboptimally, for CNS disease. However, the technological and scientific capacity to improve this situation exists.

Reformulation of drugs can improve their distribution through the BBB and BCSFB. In particular, nanoparticulate drug formulations offer promise in terms of CNS distribution. These submicrometer units may improve drug transport across the BBB by transiently increasing BBB permeability or by internalizing into brain capillary endothelial cells and thus traversing the intact BBB transvascularly (121, 122). These concepts and a description of specific carriers are presented elsewhere (123, 124). A number of examples of the exploration of nanostructured antifungal drug preparations exist. A nanosuspension formulation of amphotericin B increased AmB concentrations in mouse brains by a factor of 2.5 – 4.25 relative to LAmB (128). Amphotericin B-containing micelles, modified using a ligand of low-density lipoprotein receptor-related protein present on the BBB, have shown better BBB penetration than unmodified micelles or amphotericin B deoxycholate both *in vitro* and *in vivo* (125). Nanostructured liposomes can increase itraconazole concentrations by 2-fold in mouse brains (129). These chemical modifications represent potential avenues to maximise the utility of currently available antifungal drugs for CNS infections.

 Therapeutic drug monitoring (TDM) is a standard of care for the triazoles and 5FC regardless of the site of infection (130, 131). For CNS mycoses, antifungal TDM is even more critical to ensure adequate concentration at the effect site. Targets for TDM for efficacy are generally based on non-CNS disease. Hence, clinicians must use their judgement as to which target to aim for in TDM. From a pragmatic perspective, aiming for higher concentrations within the therapeutic range is a reasonable strategy (rather than being reassured by concentrations that are just within the therapeutic range).

**Novel antifungal drugs with potential for use as CNS agents**

 Several promising new antifungal compounds are under investigation and may hold promise as treatments for CNS disease. Viamet Pharmaceuticals (Durham, North Carolina, USA) have developed VT-1129, a quaternary azole that is efficacious in murine models of cryptococcal meningitis (132) and systemic candidiasis (133). Cidara Therapeutics (San Diego, CA, USA) are developing a novel semisynthetic echinocandin, CD101. CD101 is active against *Candida* and *Aspergillus* spp.(134), but in preclinical models displays tissue distribution patterns in keeping with currently available echinocandins, penetrating poorly into brain parenchyma (135).

 F901318 (F2G limited, Eccles, UK) is the leading compound in a novel class of antifungals, the orotomides, which block fungal pyrimidine synthesis (136). F901318 is active against *Aspergillus* spp., including resistant strains (137), as well as *Penicillium* spp., Coccidiodes immitis, H. capsulatum, Blastomyces dermatitidis, Fusarium spp., and the difficult-to-treat Scedosporium spp. (136). F901318 is detectable in brain tissue after administration to mice (136). Amplyx Pharmaceuticals (San Diego, CA, USA) have developed a novel antifungal agent, APX001, which inhibits a glycosylphosphatidylinositol-anchored fungal wall transfer protein (138). APX001 exhibits highly selective in vitro antifungal activity against *Candida* spp*.*, including strains resistant to fluconazole, Aspergillus fumigatus, A. niger, A. flavus, A. terreus, Fusarium, Pseudallescheria boydii, and S. prolificans (139, 140)*.* In a murine model of disseminated *C. auris* infection, APX001 improves survival by up to 2-fold relative to anidulafungin, with demonstrable PD effect in brain tissue (138). APX001 was granted orphan drug status by the US Food and Drug Administration in 2016 (141). Another novel first-in-class antifungal agent is SCY-078 (SCYNEXIS, Jersey City, New Jersey). SCY-078 is a semisynthetic derivative of enfumafungin and the only compound in the triterpene class of antifungals (142). It is a potent inhibitor of β-(1,3)-D-glucan synthesis in fungal cell walls and demonstrates broad spectrum activity against *Candida* spp. (143), including *C. auris* (144) and *Aspergillus* spp. (145) *in vitro*. Preclinical *in vivo* models using SCY-078 for treatment of disseminated candidiasis have demonstrated promising potency and PK (142).

**New technologies for the assessment of CNS PK of antifungals**

Historically, studies assessing the CNS PK of antifungals *in vivo* have taken brain tissue homogenates and measured drug using LC-MS, as demonstrated in this review. Modern techniques enable more refined CNS PK models including information about the spatial distribution of drug within tissues. We have seen that PK differences in various brain compartments can be assessed using microdialysis. This technique measures drug penetration into fluid compartments of the CNS using a dialysis probe to detect free drug in the cerebral interstitial fluid. The use of microdialysis in humans is limited to intraoperative settings and the technique may be unsuitable for the quantification of highly protein-bound or lipophilic drugs (146). PET scanning and MR spectroscopy are non-invasive techniques that can estimate drug distribution into the human brain by detecting radiolabelled molecules. PET can provide longitudinal trends in drug distribution but its use is limited by cost, the inability to distinguish parent compounds from metabolites, and the requirement for radioactive labelling (17, 147, 148).

Matrix-Assisted Laser Desorption and Ionisation - Mass Spectroscopy Imaging (MALDI-MSI) does not require radiolabelling and can provide rapid, highly resolved spatial drug distribution data. In murine brains, MALDI-MSI has been employed to quantify a number of antibiotics including gatifloxacin (149), doxycycline (150), pretomanid (151) and rifampicin (152) to the subcompartmental level. Serial tissue sections taken over time can yield longitudinal data. The technique has not yet been applied to assess the CNS distribution of antifungal drugs. To do so could elucidate information regarding mechanisms of drug action, distribution and inter-subject variability in CNS drug penetration. This would enable precision spatial PK modelling with vastly enhanced translational utility from laboratory animals to humans and from plasma drug levels to those in the CNS.

**Declaration of interests**

William Hope holds or has recently held research grants with F2G, AiCuris, Astellas Pharma, Spero Therapeutics, Matinas Biosciences, Antabio, Amplyx, Allecra and Pfizer. He holds awards from the National Institutes of Health, Medical Research Council, National Institute of Health Research, and the European Commission (FP7 and IMI). WH has received personal fees in his capacity as a consultant for F2G, Amplyx, Ausperix, Spero Therapeutics, Medicines Company, Gilead and Basilea. WH is Medical Guideline Director for the European Society of Clinical Microbiology and Infectious Diseases, and an Ordinary Council Member for the British Society of Antimicrobial Chemotherapy.

**Funding statement**

Katharine Stott is funded by the Wellcome Trust [grant number 203919/Z/16/Z].

**Box 1: Desirable information for the accurate setting of PK-PD targets for fungal CNS infection**

1. Principal PK parameter of interest, depending on whether drug activity is determined by maximum concentration (Cmax), area under the curve (AUC), or time above a given therapeutic threshold.
2. Drug susceptibility/ minimum inhibitory concentration (MIC) of the infecting fungus.
3. Magnitude of the PK-PD index required, in terms of the unbound concentration of drug that is sufficient to exert PD effect on the target organism.
4. Histopathological site(s) of disease within the CNS.
5. Rate constant describing the movement of drug into the CNS from the circulation.
6. Rate constant(s) describing the movement of drug within CNS compartments of interest.
7. Rate constant(s) describing the clearance of drug from the target site.
8. Rate constant(s) describing the clearance of drug from the body.
9. Time-dependent differences in plasma and tissue drug concentrations (hysteresis).
10. PK parameters associated with toxicity in measurable physiological compartments (usually the systemic circulation).
11. Clinical and physiological covariates that influence the PK of the drug in question.
12. Population-level variability in each of these factors.

**Table 1**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Fungal species** | **Clinical setting** | **Predilection for CNS involvement in invasive disease** a § | **Pathophysiological manifestations** | **Mortality with treatment (%)** | **Antifungal drug options** | **References**  |
| *Cryptococcus* spp.*C. neoformans* | Advanced HIV infection, defective cellular immunity, corticosteroid therapy | ++++ | Subacute – chronic meningitisCerebral abscesses/ cryptococcomasCerebral infarctionRaised intracranial pressure | 24 – 58 | Amphotericin BFlucytosineHigh-dose fluconazole | (32, 33, 93, 153-158) |
| *C. gattii* | Normal hosts mainly in restricted geographical locations (Australia, Southern California, British Columbia, Washington State), solid organ transplant recipients | ++++ | MeningitisCryptococcomasRaised intracranial pressure | 0 – 13 | (159-161) |
| *Coccidioides**C. immitis**C*. *posadasii* | HIV infection, steroid therapy, travel to southwest USA, Central and South America | +++ | Subacute – chronic meningitis (?basilar)Cerebral abscessesCerebral vasculitisEncephalitis | ~30 | FluconazoleItraconazoleVoriconazoleIntrathecal amphotericin B | (37, 162-166) |
| *Cladosporium* spp. | Immunodeficiency, immunosuppression, normal host, trauma | +++ | Intracerebral abscessesMeningitisEncephalitis | 29-50 | Amphotericin B in combination with vori-, itra- or posaconazole | (167-170) |
| *Aspergillus* spp. | Neutropenia, advanced HIV infection, haematopoetic stem-cell transplantation, corticosteroid therapy, chronic granulomatous disease | ++ | Cerebrovascular aspergillosis with cortical and subcortical infarction and haemorrhageAbscessesRarely: chronic meningitis  | 62 – 90 | VoriconazoleAmphotericin BIsavuconazoleConsider: High dose adjunctive echinocandins | (171-174) |
| *Scedosporium/ Pseudallescheria* spp. | NeutropeniaNear-drowning | ++ | Chronic meningitisCerebral abscess | 79 – 100\* | *S. apiospermum*: voriconazole monotherapy*S. prolificans:* voriconazole + terbinafine +/- echinocandin | (175-179) |
| *Candida* spp. | Prematurity, immunosuppression for haematological malignancies/ transplantation, invasive devices, burns, chronic granulomatous disease, advanced HIV infection | + | Subacute meningitisMultiple micro-abscesses Macroabscesses | 10 – 53 | Amphotericin BFlucytosineFluconazoleVoriconazole in fluconazole-resistant disease | (180-184) |
| *Histoplasma capsulatum* | Advanced HIV infectionPrimary or iatrogenic immunosuppressionSteroid therapySolid organ transplant recipients | + | Meningitis Cerebral mass lesionsDiffuse encephalitisRaised intracranial pressureCerebral vasculitis | 20 – 40 | Amphotericin BItraconazoleFluconazole Salvage therapy (limited data):PosaconazoleIsavuconazoleVoriconazole | (81, 185-190) |
| *Blastomycosis dermatitidis* | Normal host mainly in restricted geographical locations (southeastern, south central and midwestern USA, St Lawrence river) | + | Chronic meningitisCerebral abscesses/ blastomycomas | ~18 | Amphotericin BStep down to azole:VoriconazoleFluconazoleItraconazole | (94, 175, 191, 192) |
| Mucormycosis/ Zygomycetes | Diabetes mellitusHaematological malignancy | + | Rhino-orbital-cerebral infectionCerebral abscesses | 25 – 62 | Amphotericin BStep down to azole:PosaconazoleIsavuconazole | (95, 193, 194) |
| *Sporothrix schenckii* | HIV infectionAlcoholismEnvironmental exposure | + | Chronic meningitisEncephalitisHydrocephalus | 30-90 | Amphotericin BStep down:Itraconazole | (96, 175) |
| *Paracoccidioides* *brasiliensis* | Environmental exposure, Latin AmericaMale predominance (~23:1) | +/- | Intracerebral abscessesSpinal cord involvementHydrocephalus | ~17 | Trimethoprim-sulfamethoxazoleVoriconazoleItraconazole | (175, 195-197) |
| *Trichosporon* spp. | Haematological malignancy, HIV infection, extensive burns, intravenous catheters, heart valve surgery | No data | Intracerebral abscessesMeningitis | 70-80 | Amphotericin BVoriconazoleItraconazolePosaconazoleIsavuconazole | (198, 199) |

\* Disseminated infection

a From (200)

§ Key: ++++, very common, through +/-, very rare

**Table 2**

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Drug | Molecular weight (g/mol)a  | Particle size (µm)b | LogP a  | LogD at pH 7.4 (indicative of lipophilicity)b | Plasma protein binding (%)c | Efflux pump affinity (P-gp substrate) | Correlation of measurable CNS concentration with biological activity | References |
| Fluconazole | 309 |  | 2.17 | 0.5 | 10 | No | Good | (201-203) |
| Itraconazole | 705 |  | 6.99 | 4.9 | 98 | Yes | Poor | (202, 203) |
| Voriconazole | 349 |  | 2.56 | 2.1 | 58 | No | Good/ variable | (203) |
| Posaconazole | 700 |  | 6.1 | 4.4 | 99 | Yes | Poor | (203) |
| Isavuconazole | 718 |  | -3.33 | 3.6 | 99 | No | Good | (204, 205) |
| DAmB | 924 | <0.4 | 0.95 | -2.8 | >95 | No | Poor | (206, 207) |
| LAmB | 924 | 0.05 - 0.08 | 0.95 | -2.8 | >95 | Contentious | Poor | (14, 15, 207) |
| ABLC | 924 | 1.6 – 11 | 0.95 | -2.8 | >95 | No | Poor | (3, 207) |
| ABCD | 924 | 0.12 – 0.14 | 0.95 | -2.8 | >95 | No | Poor | (3, 207) |
| 5FC | 120 |  | -0.89 | -2.34 | 5 | No | Good | (3, 202) |
| Caspofungin | 1093 |  | -2.8 | -3.88 | 98 | No | Good | (208, 209) |
| Micafungin | 1140 |  | -3.8 | -1.62 | 98 | No | Good | (208, 209) |
| Anidulafungin | 1291 |  | 0.21 | -3.32 | 98 | No | Good | (116, 209) |

a From reference (3)

b From reference (2, 210)

c From references (3, 203)

**References**

1. Brown GD, Denning DW, Gow NAR, Levitz SM, Netea MG, White TC. Hidden Killers: Human Fungal Infections. Science Translational Medicine. 2012;4(165):165rv13-rv13.

2. Felton T, Troke PF, Hope WW. Tissue penetration of antifungal agents. Clin Microbiol Rev. 2014;27(1):68-88.

3. Kethireddy S, Andes D. CNS pharmacokinetics of antifungal agents. Expert opinion on drug metabolism & toxicology. 2007;3(4):573-81.

4. de Lange ECM. The mastermind approach to CNS drug therapy: translational prediction of human brain distribution, target site kinetics, and therapeutic effects. Fluids and Barriers of the CNS. 2013;10(1):12.

**\*\* Introduction to CNS PK-PD including sources of associated variability.**

5. Nau R, Sorgel F, Eiffert H. Penetration of drugs through the blood-cerebrospinal fluid/blood-brain barrier for treatment of central nervous system infections. Clin Microbiol Rev. 2010;23(4):858-83.

6. Di Paolo A, Gori G, Tascini C, Danesi R, Del Tacca M. Clinical pharmacokinetics of antibacterials in cerebrospinal fluid. Clin Pharmacokinet. 2013;52(7):511-42.

7. Fong IW, Tomkins KB. Penetration of ceftazidime into the cerebrospinal fluid of patients with and without evidence of meningeal inflammation. Antimicrobial Agents and Chemotherapy. 1984;26(1):115-6.

8. Pardridge WM. Blood-brain barrier drug targeting: the future of brain drug development. Mol Interv. 2003;3(2):90-105, 51.

9. De Lange ECM, Danhof M. Considerations in the use of cerebrospinal fluid pharmacokinetic to predict brain target concentrations in the clinical setting. Implications of the barriers between blood and brain. Clin Pharmacokinet. 2002;41.

10. Andes DR, Craig WA. Pharmacokinetics and pharmacodynamics of antibiotics in meningitis. Infect Dis Clin North Am. 1999;13(3):595-618.

11. Atkinson F, Cole S, Green C, Van de Waterbeemd H. Lipophilicity and other parameters affecting brain penetration. Current Medicinal Chemistry-Central Nervous System Agents. 2002;2(3):229-40.

12. Yamamoto Y, Danhof M, de Lange ECM. Microdialysis: the Key to Physiologically Based Model Prediction of Human CNS Target Site Concentrations. AAPS J. 2017;19(4):891-909.

13. Lempers VJC, van den Heuvel JJMW, Russel FGM, Aarnoutse RE, Burger DM, Brüggemann RJ, et al. Inhibitory Potential of Antifungal Drugs on ATP-Binding Cassette Transporters P-Glycoprotein, MRP1 to MRP5, BCRP, and BSEP. Antimicrobial Agents and Chemotherapy. 2016;60(6):3372-9.

14. Wu JQ, Shao K, Wang X, Wang RY, Cao YH, Yu YQ, et al. In vitro and in vivo evidence for amphotericin B as a P-glycoprotein substrate on the blood-brain barrier. Antimicrob Agents Chemother. 2014;58(8):4464-9.

15. Stevens DA, Clemons KV, Martinez M, Chen V. The Brain, Amphotericin B, and P-Glycoprotein. Antimicrobial Agents and Chemotherapy. 2015;59(2):1386.

16. Stott KE, Hope WW. Therapeutic drug monitoring for invasive mould infections and disease: pharmacokinetic and pharmacodynamic considerations. J Antimicrob Chemother. 2017;72(suppl\_1):i12-i8.

17. Srinivas N, Maffuid K, Kashuba ADM. Clinical Pharmacokinetics and Pharmacodynamics of Drugs in the Central Nervous System. Clinical Pharmacokinetics. 2018.

**\*\* Excellent introduction to factors of relevance for understanding CNS PK-PD**

18. Hope WW, Mickiene D, Petraitis V, Petraitiene R, Kelaher AM, Hughes JE, et al. The Pharmacokinetics and Pharmacodynamics of Micafungin in Experimental Hematogenous Candida Meningoencephalitis: Implications for Echinocandin Therapy in Neonates. The Journal of Infectious Diseases. 2008;197(1):163-71.

**\* Demonstrates the potential of PK-PD modelling to predict clinical outcomes**

19. Warn PA, Livermore J, Howard S, Felton TW, Sharp A, Gregson L, et al. Anidulafungin for neonatal hematogenous Candida meningoencephalitis: identification of candidate regimens for humans using a translational pharmacological approach. Antimicrob Agents Chemother. 2012;56(2):708-14.

20. Yamamoto Y, Välitalo PA, van den Berg DJ, Hartman R, van den Brink W, Wong YC, et al. A Generic Multi-Compartmental CNS Distribution Model Structure for 9 Drugs Allows Prediction of Human Brain Target Site Concentrations. Pharm Res. 2017;34(2):333-51.

21. Yamamoto Y, Valitalo PA, Wong YC, Huntjens DR, Proost JH, Vermeulen A, et al. Prediction of human CNS pharmacokinetics using a physiologically-based pharmacokinetic modeling approach. Eur J Pharm Sci. 2018;112:168-79.

22. Alqahtani S, Kaddoumi A. Development of a Physiologically Based Pharmacokinetic/Pharmacodynamic Model to Predict the Impact of Genetic Polymorphisms on the Pharmacokinetics and Pharmacodynamics Represented by Receptor/Transporter Occupancy of Central Nervous System Drugs. Clin Pharmacokinet. 2016;55(8):957-69.

23. Tucker RM, Williams PL, Arathoon EG, Levine BE, Hartstein AI, Hanson LH, et al. Pharmacokinetics of fluconazole in cerebrospinal fluid and serum in human coccidioidal meningitis. Antimicrobial Agents and Chemotherapy. 1988;32(3):369-73.

24. Thaler F, Bernard B, Tod M, Jedynak CP, Petitjean O, Derome P, et al. Fluconazole penetration in cerebral parenchyma in humans at steady state. Antimicrobial Agents and Chemotherapy. 1995;39(5):1154-6.

25. Fischman AJ, Alpert NM, Livni E, Ray S, Sinclair I, Callahan RJ, et al. Pharmacokinetics of 18F-labeled fluconazole in healthy human subjects by positron emission tomography. Antimicrob Agents Chemother. 1993;37(6):1270-7.

26. Henry ME, Bolo NR, Zuo CS, Villafuerte RA, Cayetano K, Glue P, et al. Quantification of brain voriconazole levels in healthy adults using fluorine magnetic resonance spectroscopy. Antimicrob Agents Chemother. 2013;57(11):5271-6.

27. Yang H, Wang Q, Elmquist WF. Fluconazole distribution to the brain: a crossover study in freely-moving rats using in vivo microdialysis. Pharm Res. 1996;13(10):1570-5.

**\* Elegant demonstration of the use of microdialysis to assess CNS drug distribution**

28. Arndt CA, Walsh TJ, McCully CL, Balis FM, Pizzo PA, Poplack DG. Fluconazole penetration into cerebrospinal fluid: implications for treating fungal infections of the central nervous system. Journal of Infectious Diseases. 1988;157(1):178-80.

29. Madu A, Cioffe C, Mian U, Burroughs M, Tuomanen E, Mayers M, et al. Pharmacokinetics of fluconazole in cerebrospinal fluid and serum of rabbits: validation of an animal model used to measure drug concentrations in cerebrospinal fluid. Antimicrob Agents Chemother. 1994;38(9):2111-5.

30. Brammer KW, Farrow PR, Faulkner JK. Pharmacokinetics and tissue penetration of fluconazole in humans. Rev Infect Dis. 1990;12 Suppl 3:S318-26.

31. WHO. Rapid advice: Diagnosis, prevention and management of cryptococcal disease in HIV-infected adults, adolescents and children.; 2011.

32. Longley N, Muzoora C, Taseera K, Mwesigye J, Rwebembera J, Chakera A, et al. Dose response effect of high-dose fluconazole for HIV-associated cryptococcal meningitis in southwestern Uganda. Clin Infect Dis. 2008;47(12):1556-61.

33. Nussbaum JC, Jackson A, Namarika D, Phulusa J, Kenala J, Kanyemba C, et al. Combination flucytosine and high-dose fluconazole compared with fluconazole monotherapy for the treatment of cryptococcal meningitis: a randomized trial in Malawi. Clin Infect Dis. 2010;50(3):338-44.

34. Bicanic T, Meintjes G, Wood R, Hayes M, Rebe K, Bekker L-G, et al. Fungal Burden, Early Fungicidal Activity, and Outcome in Cryptococcal Meningitis in Antiretroviral-Naive or Antiretroviral-Experienced Patients Treated with Amphotericin B or Fluconazole. Clinical Infectious Diseases. 2007;45(1):76-80.

35. Brouwer AE, Rajanuwong A, Chierakul W, Griffin GE, Larsen RA, White NJ, et al. Combination antifungal therapies for HIV-associated cryptococcal meningitis: a randomised trial. Lancet. 2004;363(9423):1764-7.

36. Joint\_Formulary\_Committee. British National Formulary (online) London: BMJ Group and Pharmaceutical Press; 2016 [

37. Galgiani JN, Ampel NM, Blair JE, Catanzaro A, Geertsma F, Hoover SE, et al. 2016 Infectious Diseases Society of America (IDSA) Clinical Practice Guideline for the Treatment of Coccidioidomycosis. Clin Infect Dis. 2016;63(6):e112-46.

38. Sorensen KN, Sobel RA, Clemons KV, Pappagianis D, Stevens DA, Williams PL. Comparison of Fluconazole and Itraconazole in a Rabbit Model of Coccidioidal Meningitis. Antimicrobial Agents and Chemotherapy. 2000;44(6):1512-7.

39. J H, M M, W M, J M, K L, A VP, et al. The pharmacokinetics of itraconazole in animals and man: an overview. In: RA F, editor. Recent trends in the discovery, development and evaluation of antifungal agents. Barcelona, Spain: J R Prous; 1987. p. 223 - 59.

40. Perfect JR, Savani DV, Durack DT. Comparison of itraconazole and fluconazole in treatment of cryptococcal meningitis and candida pyelonephritis in rabbits. Antimicrob Agents Chemother. 1986;29(4):579-83.

41. Poirier JM, Cheymol G. Optimisation of itraconazole therapy using target drug concentrations. Clin Pharmacokinet. 1998;35(6):461-73.

42. Miyama T, Takanaga H, Matsuo H, Yamano K, Yamamoto K, Iga T, et al. P-Glycoprotein-Mediated Transport of Itraconazole across the Blood-Brain Barrier. Antimicrobial Agents and Chemotherapy. 1998;42(7):1738-44.

43. Haynes RR, Connolly PA, Durkin MM, LeMonte AM, Smedema ML, Brizendine E, et al. Antifungal therapy for central nervous system histoplasmosis, using a newly developed intracranial model of infection. Journal of Infectious Diseases. 2002;185(12):1830-2.

44. Imbert F, Jardin M, Fernandez C, Gantier JC, Dromer F, Baron G, et al. Effect of efflux inhibition on brain uptake of itraconazole in mice infected with Cryptococcus neoformans. Drug Metab Dispos. 2003;31(3):319-25.

45. Chiller TM, Sobel RA, Luque JC, Clemons KV, Stevens DA. Efficacy of amphotericin B or itraconazole in a murine model of central nervous system Aspergillus infection. Antimicrob Agents Chemother. 2003;47(2):813-5.

46. Pfaller MA, Espinel-Ingroff A, Canton E, Castanheira M, Cuenca-Estrella M, Diekema DJ, et al. Wild-type MIC distributions and epidemiological cutoff values for amphotericin B, flucytosine, and itraconazole and Candida spp. as determined by CLSI broth microdilution. J Clin Microbiol. 2012;50(6):2040-6.

47. Chang LW, Phipps WT, Kennedy GE, Rutherford GW. Antifungal interventions for the primary prevention of cryptococcal disease in adults with HIV. Cochrane Database Syst Rev. 2005(3):Cd004773.

48. Nelson MR, Bower M, Smith D, Reed C, Shanson D, Gazzard B. The value of serum cryptococcal antigen in the diagnosis of cryptococcal infection in patients infected with the human immunodeficiency virus. J Infect. 1990;21(2):175-81.

49. Denning DW, Tucker RM, Hanson LH, Hamilton JR, Stevens DA. Itraconazole therapy for cryptococcal meningitis and cryptococcosis. Archives of Internal Medicine. 1989;149(10):2301-8.

50. Sharkey PK, Rinaldi MG, Dunn JF, Hardin TC, Fetchick RJ, Graybill JR. High-dose itraconazole in the treatment of severe mycoses. Antimicrob Agents Chemother. 1991;35(4):707-13.

51. Saag MS, Cloud GA, Graybill JR, Sobel JD, Tuazon CU, Johnson PC, et al. A comparison of itraconazole versus fluconazole as maintenance therapy for AIDS-associated cryptococcal meningitis. National Institute of Allergy and Infectious Diseases Mycoses Study Group. Clin Infect Dis. 1999;28(2):291-6.

52. McKinsey DS, Wheat LJ, Cloud GA, Pierce M, Black JR, Bamberger DM, et al. Itraconazole prophylaxis for fungal infections in patients with advanced human immunodeficiency virus infection: randomized, placebo-controlled, double-blind study. National Institute of Allergy and Infectious Diseases Mycoses Study Group. Clin Infect Dis. 1999;28(5):1049-56.

53. Lestner J, Hope WW. Itraconazole: an update on pharmacology and clinical use for treatment of invasive and allergic fungal infections. Expert Opin Drug Metab Toxicol. 2013;9(7):911-26.

54. Lutsar I, Roffey S, Troke P. Voriconazole concentrations in the cerebrospinal fluid and brain tissue of guinea pigs and immunocompromised patients. Clin Infect Dis. 2003;37(5):728-32.

55. Elter T, Sieniawski M, Gossmann A, Wickenhauser C, Schroder U, Seifert H, et al. Voriconazole brain tissue levels in rhinocerebral aspergillosis in a successfully treated young woman. Int J Antimicrob Agents. 2006;28(3):262-5.

56. Denes E, Pichon N, Debette-Gratien M, Bouteille B, Gaulier JM. Pharmacokinetics of voriconazole in the cerebrospinal fluid of an immunocompromised patient with a brain abscess due to Aspergillus fumigatus. Clin Infect Dis. 2004;39(4):603-4.

57. Walsh TJ, Karlsson MO, Driscoll T, Arguedas AG, Adamson P, Saez-Llorens X, et al. Pharmacokinetics and safety of intravenous voriconazole in children after single- or multiple-dose administration. Antimicrob Agents Chemother. 2004;48(6):2166-72.

58. Hope WW. Population pharmacokinetics of voriconazole in adults. Antimicrob Agents Chemother. 2012;56(1):526-31.

59. Friberg LE, Ravva P, Karlsson MO, Liu P. Integrated population pharmacokinetic analysis of voriconazole in children, adolescents, and adults. Antimicrob Agents Chemother. 2012;56(6):3032-42.

**\* Demonstration of the utility of PK-PD modelling in quantifying PK variability in different populations**

60. Kadam RS, Van Den Anker JN. Pediatric Clinical Pharmacology of Voriconazole: Role of Pharmacokinetic/Pharmacodynamic Modeling in Pharmacotherapy. Clinical Pharmacokinetics. 2016:1-13.

61. Luong M-L, Al-Dabbagh M, Groll AH, Racil Z, Nannya Y, Mitsani D, et al. Utility of voriconazole therapeutic drug monitoring: a meta-analysis. Journal of Antimicrobial Chemotherapy. 2016;71(7):1786-99.

62. Patterson TF, Thompson IIIGR, Denning DW, Fishman JA, Hadley S, Herbrecht R, et al. Practice Guidelines for the Diagnosis and Management of Aspergillosis: 2016 Update by the Infectious Diseases Society of America. Clinical Infectious Diseases. 2016;63(4):e1-e60.

63. Nagappan V, Deresinski S. Posaconazole: A Broad-Spectrum Triazole Antifungal Agent. Clinical Infectious Diseases. 2007;45(12):1610-7.

64. Sansone-Parsons A, Krishna G, Simon J, Soni P, Kantesaria B, Herron J, et al. Effects of age, gender, and race/ethnicity on the pharmacokinetics of posaconazole in healthy volunteers. Antimicrob Agents Chemother. 2007;51(2):495-502.

65. Ullmann AJ, Cornely OA, Burchardt A, Hachem R, Kontoyiannis DP, Topelt K, et al. Pharmacokinetics, safety, and efficacy of posaconazole in patients with persistent febrile neutropenia or refractory invasive fungal infection. Antimicrob Agents Chemother. 2006;50(2):658-66.

66. Gubbins PO, Krishna G, Sansone-Parsons A, Penzak SR, Dong L, Martinho M, et al. Pharmacokinetics and safety of oral posaconazole in neutropenic stem cell transplant recipients. Antimicrob Agents Chemother. 2006;50(6):1993-9.

67. Petitcollin A, Boglione-Kerrien C, Tron C, Nimubona S, Lalanne S, Lemaitre F, et al. Population Pharmacokinetics of Posaconazole Tablets and Monte Carlo Simulations To Determine whether All Patients Should Receive the Same Dose. Antimicrob Agents Chemother. 2017;61(11).

68. Durani U, Tosh PK, Barreto JN, Estes LL, Jannetto PJ, Tande AJ. Retrospective Comparison of Posaconazole Levels in Patients Taking the Delayed-Release Tablet versus the Oral Suspension. Antimicrob Agents Chemother. 2015;59(8):4914-8.

69. Cumpston A, Caddell R, Shillingburg A, Lu X, Wen S, Hamadani M, et al. Superior Serum Concentrations with Posaconazole Delayed-Release Tablets Compared to Suspension Formulation in Hematological Malignancies. Antimicrob Agents Chemother. 2015;59(8):4424-8.

70. Krishna G, Ma L, Martinho M, O'Mara E. Single-Dose Phase I Study To Evaluate the Pharmacokinetics of Posaconazole in New Tablet and Capsule Formulations Relative to Oral Suspension. Antimicrobial Agents and Chemotherapy. 2012;56(8):4196-201.

71. Kersemaekers WM, van Iersel T, Nassander U, O'Mara E, Waskin H, Caceres M, et al. Pharmacokinetics and Safety Study of Posaconazole Intravenous Solution Administered Peripherally to Healthy Subjects. Antimicrobial Agents and Chemotherapy. 2015;59(2):1246-51.

72. Calvo E, Pastor FJ, Rodríguez MM, Pujol I, Guarro J. Antifungal Therapy in a Murine Model of Disseminated Infection by Cryptococcus gattii. Antimicrobial Agents and Chemotherapy. 2010;54(10):4074-7.

73. Calvo E, Pastor FJ, Rodríguez MM, Mayayo E, Salas V, Guarro J. Murine Model of a Disseminated Infection by the Novel Fungus Fonsecaea monophora and Successful Treatment with Posaconazole. Antimicrobial Agents and Chemotherapy. 2010;54(2):919-23.

74. Calcagno A, Baietto L, De Rosa FG, Tettoni MC, Libanore V, Bertucci R, et al. Posaconazole cerebrospinal concentrations in an HIV-infected patient with brain mucormycosis. J Antimicrob Chemother. 2011;66(1):224-5.

75. Ruping MJ, Albermann N, Ebinger F, Burckhardt I, Beisel C, Muller C, et al. Posaconazole concentrations in the central nervous system. J Antimicrob Chemother. 2008;62(6):1468-70.

76. Reinwald M, Uharek L, Lampe D, Grobosch T, Thiel E, Schwartz S. Limited penetration of posaconazole into cerebrospinal fluid in an allogeneic stem cell recipient with invasive pulmonary aspergillosis. Bone Marrow Transplant. 2009;44(4):269-70.

77. Anstead GM, Corcoran G, Lewis J, Berg D, Graybill JR. Refractory coccidioidomycosis treated with posaconazole. Clin Infect Dis. 2005;40(12):1770-6.

78. Vehreschild JJ, Birtel A, Vehreschild MJGT, Liss B, Farowski F, Kochanek M, et al. Mucormycosis treated with posaconazole: review of 96 case reports. Critical Reviews in Microbiology. 2013;39(3):310-24.

79. Simmonds L, Mitchell S, White B, Crusz SA, Denning D. Aspergillus niger infection in an immunosuppressed patient confined solely to the brain. BMJ Case Rep. 2017;2017.

80. Walsh TJ, Raad I, Patterson TF, Chandrasekar P, Donowitz GR, Graybill R, et al. Treatment of invasive aspergillosis with posaconazole in patients who are refractory to or intolerant of conventional therapy: an externally controlled trial. Clin Infect Dis. 2007;44(1):2-12.

81. Restrepo A, Tobon A, Clark B, Graham DR, Corcoran G, Bradsher RW, et al. Salvage treatment of histoplasmosis with posaconazole. J Infect. 2007;54(4):319-27.

82. Schmitt-Hoffmann AH, Kato K, Townsend R, Potchoiba MJ, Hope WW. Tissue Distribution and Elimination of Isavuconazole Following Single and Repeat Oral-Dose Administration of Isavuconazonium Sulfate to Rats. 2017.

83. Wiederhold NP, Kovanda L, Najvar LK, Bocanegra R, Olivo M, Kirkpatrick WR, et al. Isavuconazole Is Effective for the Treatment of Experimental Cryptococcal Meningitis. Antimicrob Agents Chemother. 2016;60(9):5600-3.

84. Schwartz. ASM Microbe; Boston, Massachusetts: American Society for Microbiology; 2016.

85. Hamill RJ. Amphotericin B Formulations: A Comparative Review of Efficacy and Toxicity. Drugs. 2013;73(9):919-34.

86. Groll AH, Giri N, Petraitis V, Petraitiene R, Candelario M, Bacher JS, et al. Comparative efficacy and distribution of lipid formulations of amphotericin B in experimental Candida albicans infection of the central nervous system. J Infect Dis. 2000;182(1):274-82.

87. Lee JW, Amantea MA, Francis PA, Navarro EE, Bacher J, Pizzo PA, et al. Pharmacokinetics and safety of a unilamellar liposomal formulation of amphotericin B (AmBisome) in rabbits. Antimicrob Agents Chemother. 1994;38(4):713-8.

88. Pyrgos V, Mickiene D, Sein T, Cotton M, Fransesconi A, Mizrahi I, et al. Effects of immunomodulatory and organism-associated molecules on the permeability of an in vitro blood-brain barrier model to amphotericin B and fluconazole. Antimicrob Agents Chemother. 2010;54(3):1305-10.

89. Lestner J, McEntee L, Johnson A, Livermore J, Whalley S, Schwartz J, et al. Experimental Models of Short Courses of Liposomal Amphotericin B for Induction Therapy for Cryptococcal Meningitis. Antimicrob Agents Chemother. 2017.

90. Livermore J, Howard SJ, Sharp AD, Goodwin J, Gregson L, Felton T, et al. Efficacy of an abbreviated induction regimen of amphotericin B deoxycholate for cryptococcal meningoencephalitis: 3 days of therapy is equivalent to 14 days. MBio. 2014;5(1):e00725-13.

**\* Important preclinical study demonstrating the efficacy of abbreviated regimens of liposomal amphotericin B.**

91. Strenger V, Meinitzer A, Donnerer J, Hofer N, Dornbusch HJ, Wanz U, et al. Amphotericin B transfer to CSF following intravenous administration of liposomal amphotericin B. J Antimicrob Chemother. 2014;69(9):2522-6.

92. Vogelsinger H, Weiler S, Djanani A, Kountchev J, Bellmann-Weiler R, Wiedermann CJ, et al. Amphotericin B tissue distribution in autopsy material after treatment with liposomal amphotericin B and amphotericin B colloidal dispersion. J Antimicrob Chemother. 2006;57(6):1153-60.

93. Bicanic T, Wood R, Meintjes G, Rebe K, Brouwer A, Loyse A, et al. High-dose amphotericin B with flucytosine for the treatment of cryptococcal meningitis in HIV-infected patients: a randomized trial. Clin Infect Dis. 2008;47(1):123-30.

94. Bariola JR, Perry P, Pappas PG, Proia L, Shealey W, Wright PW, et al. Blastomycosis of the Central Nervous System: A Multicenter Review of Diagnosis and Treatment in the Modern Era. Clinical Infectious Diseases. 2010;50(6):797-804.

95. Roden MM, Zaoutis TE, Buchanan WL, Knudsen TA, Sarkisova TA, Schaufele RL, et al. Epidemiology and outcome of zygomycosis: a review of 929 reported cases. Clin Infect Dis. 2005;41(5):634-53.

96. Moreira JAS, Freitas DFS, Lamas CC. The impact of sporotrichosis in HIV-infected patients: a systematic review. Infection. 2015;43(3):267-76.

97. O'Connor L, Livermore J, Sharp AD, Goodwin J, Gregson L, Howard SJ, et al. Pharmacodynamics of liposomal amphotericin B and flucytosine for cryptococcal meningoencephalitis: safe and effective regimens for immunocompromised patients. J Infect Dis. 2013;208(2):351-61.

98. Brouwer AE, van Kan HJM, Johnson E, Rajanuwong A, Teparrukkul P, Wuthiekanun V, et al. Oral versus Intravenous Flucytosine in Patients with Human Immunodeficiency Virus-Associated Cryptococcal Meningitis. Antimicrobial Agents and Chemotherapy. 2007;51(3):1038-42.

99. Block ER, Bennett JE. Pharmacological studies with 5-fluorocytosine. Antimicrob Agents Chemother. 1972;1(6):476-82.

100. Andes D, van Ogtrop M. In vivo characterization of the pharmacodynamics of flucytosine in a neutropenic murine disseminated candidiasis model. Antimicrob Agents Chemother. 2000;44(4):938-42.

101. Lewis RE, Klepser ME, Pfaller MA. In vitro pharmacodynamic characteristics of flucytosine determined by time-kill methods. Diagn Microbiol Infect Dis. 2000;36(2):101-5.

102. Polak A, Scholer HJ. Mode of action of 5-fluorocytosine and mechanisms of resistance. Chemotherapy. 1975;21(3-4):113-30.

103. Normark S, Schonebeck J. In vitro studies of 5-fluorocytosine resistance in Candida albicans and Torulopsis glabrata. Antimicrob Agents Chemother. 1972;2(3):114-21.

104. Tassel D, Madoff MA. Treatment of Candida sepsis and Cryptococcus meningitis with 5-fluorocytosine: a new antifungal agent. Jama. 1968;206(4):830-2.

105. Hospenthal DR, Bennett JE. Flucytosine monotherapy for cryptococcosis. Clin Infect Dis. 1998;27(2):260-4.

106. Day JN, Chau TT, Wolbers M, Mai PP, Dung NT, Mai NH, et al. Combination antifungal therapy for cryptococcal meningitis. N Engl J Med. 2013;368(14):1291-302.

107. van der Horst CM, Saag MS, Cloud GA, Hamill RJ, Graybill JR, Sobel JD, et al. Treatment of Cryptococcal Meningitis Associated with the Acquired Immunodeficiency Syndrome. New England Journal of Medicine. 1997;337(1):15-21.

108. Bennett JE, Dismukes WE, Duma RJ, Medoff G, Sande MA, Gallis H, et al. A Comparison of Amphotericin B Alone and Combined with Flucytosine in the Treatment of Cryptoccal Meningitis. New England Journal of Medicine. 1979;301(3):126-31.

109. Medoff G, Comfort M, Kobayashi GS. Synergistic action of amphotericin B and 5-fluorocytosine against yeast-like organisms. Proc Soc Exp Biol Med. 1971;138(2):571-4.

110. Montgomerie JZ, Edwards JE, Jr., Guze LB. Synergism of amphotericin B and 5-fluorocytosine for candida species. J Infect Dis. 1975;132(1):82-6.

111. Smego RA, Jr., Perfect JR, Durack DT. Combined therapy with amphotericin B and 5-fluorocytosine for Candida meningitis. Rev Infect Dis. 1984;6(6):791-801.

112. Joint\_Formulary\_Committee. British National Formulary for Children (online) London: BMJ Group, Royal Pharmaceutical Society of Great Britain, RCPCH Publications; 2018

113. Brian Smith P, Steinbach WJ, Benjamin DK. Invasive Candida infections in the neonate. Drug Resistance Updates. 2005;8(3):147-62.

114. Schwartz S, Thiel E. Cerebral aspergillosis: tissue penetration is the key. Med Mycol. 2009;47 Suppl 1:S387-93.

115. Groll AH, Mickiene D, Petraitis V, Petraitiene R, Ibrahim KH, Piscitelli SC, et al. Compartmental pharmacokinetics and tissue distribution of the antifungal echinocandin lipopeptide micafungin (FK463) in rabbits. Antimicrob Agents Chemother. 2001;45(12):3322-7.

116. Groll AH, Mickiene D, Petraitiene R, Petraitis V, Lyman CA, Bacher JS, et al. Pharmacokinetic and pharmacodynamic modeling of anidulafungin (LY303366): reappraisal of its efficacy in neutropenic animal models of opportunistic mycoses using optimal plasma sampling. Antimicrob Agents Chemother. 2001;45(10):2845-55.

117. Hajdu R, Thompson R, Sundelof JG, Pelak BA, Bouffard FA, Dropinski JF, et al. Preliminary animal pharmacokinetics of the parenteral antifungal agent MK-0991 (L-743,872). Antimicrob Agents Chemother. 1997;41(11):2339-44.

118. Lat A, Thompson GR, Rinaldi MG, Dorsey SA, Pennick G, Lewis JS. Micafungin Concentrations from Brain Tissue and Pancreatic Pseudocyst Fluid. Antimicrobial Agents and Chemotherapy. 2010;54(2):943-4.

119. Okugawa S, Ota Y, Tatsuno K, Tsukada K, Kishino S, Koike K. A case of invasive central nervous system aspergillosis treated with micafungin with monitoring of micafungin concentrations in the cerebrospinal fluid. Scand J Infect Dis. 2007;39(4):344-6.

120. Prabhu RM, Orenstein R. Failure of caspofungin to treat brain abscesses secondary to Candida albicans prosthetic valve endocarditis. Clin Infect Dis. 2004;39(8):1253-4.

121. Garcia-Garcia E, Andrieux K, Gil S, Couvreur P. Colloidal carriers and blood-brain barrier (BBB) translocation: a way to deliver drugs to the brain? Int J Pharm. 2005;298(2):274-92.

122. Alyautdin R, Khalin I, Nafeeza MI, Haron MH, Kuznetsov D. Nanoscale drug delivery systems and the blood–brain barrier. International Journal of Nanomedicine. 2014;9:795-811.

123. Li X, Tsibouklis J, Weng T, Zhang B, Yin G, Feng G, et al. Nano carriers for drug transport across the blood–brain barrier. Journal of Drug Targeting. 2017;25(1):17-28.

124. He Q, Liu J, Liang J, Liu X, Li W, Liu Z, et al. Towards Improvements for Penetrating the Blood–Brain Barrier—Recent Progress from a Material and Pharmaceutical Perspective. Cells. 2018;7(4):24.

**\*\* Overview of strategies to enhance CNS drug delivery**

125. Shao K, Huang R, Li J, Han L, Ye L, Lou J, et al. Angiopep-2 modified PE-PEG based polymeric micelles for amphotericin B delivery targeted to the brain. J Control Release. 2010;147(1):118-26.

126. Gershkovich P, Sivak O, Wasan EK, Magil AB, Owen D, Clement JG, et al. Biodistribution and tissue toxicity of amphotericin B in mice following multiple dose administration of a novel oral lipid-based formulation (iCo-009). J Antimicrob Chemother. 2010;65(12):2610-3.

127. Wasan EK, Gershkovich P, Zhao J, Zhu X, Werbovetz K, Tidwell RR, et al. A novel tropically stable oral amphotericin B formulation (iCo-010) exhibits efficacy against visceral Leishmaniasis in a murine model. PLoS neglected tropical diseases. 2010;4(12):e913.

128. Lemke A, Kiderlen AF, Petri B, Kayser O. Delivery of amphotericin B nanosuspensions to the brain and determination of activity against Balamuthia mandrillaris amebas. Nanomedicine. 2010;6(4):597-603.

129. Lim WM, Rajinikanth PS, Mallikarjun C, Kang YB. Formulation and delivery of itraconazole to the brain using a nanolipid carrier system. International Journal of Nanomedicine. 2014;9:2117-26.

130. Pascual A, Calandra T, Bolay S, Buclin T, Bille J, Marchetti O. Voriconazole therapeutic drug monitoring in patients with invasive mycoses improves efficacy and safety outcomes. Clin Infect Dis. 2008;46(2):201-11.

131. Lestner JM, Roberts SA, Moore CB, Howard SJ, Denning DW, Hope WW. Toxicodynamics of Itraconazole: Implications for Therapeutic Drug Monitoring. Clinical Infectious Diseases. 2009;49(6):928-30.

132. NP W, LK N, A A, J C. The novel fungal Cyp51 inhibitor VT-1129 demonstrates potent in vivo activity in mice against cryptococcal meningitis with a loading/maintenance dose strategy. ECCMID; Copenhagen2015.

133. Hoekstra WJ, Garvey EP, Moore WR, Rafferty SW, Yates CM, Schotzinger RJ. Design and optimization of highly-selective fungal CYP51 inhibitors. Bioorg Med Chem Lett. 2014;24(15):3455-8.

134. Pfaller MA, Messer SA, Rhomberg PR, Jones RN, Castanheira M. Activity of a long-acting echinocandin, CD101, determined using CLSI and EUCAST reference methods, against Candida and Aspergillus spp., including echinocandin- and azole-resistant isolates. J Antimicrob Chemother. 2016;71(10):2868-73.

135. Ong V, James KD, Smith S, Krishnan BR. Pharmacokinetics of the Novel Echinocandin CD101 in Multiple Animal Species. Antimicrob Agents Chemother. 2017;61(4).

136. Oliver JD, Sibley GEM, Beckmann N, Dobb KS, Slater MJ, McEntee L, et al. F901318 represents a novel class of antifungal drug that inhibits dihydroorotate dehydrogenase. Proceedings of the National Academy of Sciences. 2016;113(45):12809-14.

137. Buil JB, Rijs A, Meis JF, Birch M, Law D, Melchers WJG, et al. In vitro activity of the novel antifungal compound F901318 against difficult-to-treat Aspergillus isolates. J Antimicrob Chemother. 2017;72(9):2548-52.

138. Hager CL, Larkin EL, Long L, Abidi FZ, Shaw KJ, Ghannoum MA. In vitro and in vivo Evaluation of the Antifungal Activity of APX001A/APX001 Against Candida auris. Antimicrob Agents Chemother. 2018.

139. Miyazaki M, Horii T, Hata K, Watanabe NA, Nakamoto K, Tanaka K, et al. In vitro activity of E1210, a novel antifungal, against clinically important yeasts and molds. Antimicrob Agents Chemother. 2011;55(10):4652-8.

140. Watanabe NA, Miyazaki M, Horii T, Sagane K, Tsukahara K, Hata K. E1210, a new broad-spectrum antifungal, suppresses Candida albicans hyphal growth through inhibition of glycosylphosphatidylinositol biosynthesis. Antimicrob Agents Chemother. 2012;56(2):960-71.

141. AmplyxPharmaceuticals. Amplyx Pharmaceuticals Announces Qualified Infectious Disease Product Designation for APX001 in Three Life-threatening Fungal Infections 2016 [Available from: <http://amplyx.com/amplyx-pharmaceuticals-announces-qualified-infectious-disease-product-designation-for-apx001-in-three-life-threatening-fungal-infections/>.]

142. Wring SA, Randolph R, Park S, Abruzzo G, Chen Q, Flattery A, et al. Preclinical Pharmacokinetics and Pharmacodynamic Target of SCY-078, a First-in-Class Orally Active Antifungal Glucan Synthesis Inhibitor, in Murine Models of Disseminated Candidiasis. Antimicrobial Agents and Chemotherapy. 2017;61(4).

143. Pfaller MA, Messer SA, Motyl MR, Jones RN, Castanheira M. Activity of MK-3118, a new oral glucan synthase inhibitor, tested against Candida spp. by two international methods (CLSI and EUCAST). J Antimicrob Chemother. 2013;68(4):858-63.

144. Berkow EL, Angulo D, Lockhart SR. In Vitro Activity of a Novel Glucan Synthase Inhibitor, SCY-078, against Clinical Isolates of Candida auris. Antimicrobial Agents and Chemotherapy. 2017;61(7).

145. Pfaller MA, Messer SA, Motyl MR, Jones RN, Castanheira M. In vitro activity of a new oral glucan synthase inhibitor (MK-3118) tested against Aspergillus spp. by CLSI and EUCAST broth microdilution methods. Antimicrob Agents Chemother. 2013;57(2):1065-8.

146. Shannon RJ, Carpenter KLH, Guilfoyle MR, Helmy A, Hutchinson PJ. Cerebral microdialysis in clinical studies of drugs: pharmacokinetic applications. Journal of Pharmacokinetics and Pharmacodynamics. 2013;40(3):343-58.

147. Varnas K, Varrone A, Farde L. Modeling of PET data in CNS drug discovery and development. J Pharmacokinet Pharmacodyn. 2013;40(3):267-79.

148. Neuwelt EA, Abbott NJ, Abrey L, Banks WA, Blakley B, Davis T, et al. Strategies to advance translational research into brain barriers. Lancet Neurol. 2008;7.

149. Shobo A, Baijnath S, Bratkowska D, Naiker S, Somboro AM, Bester LA, et al. MALDI MSI and LC-MS/MS: Towards preclinical determination of the neurotoxic potential of fluoroquinolones. Drug Test Anal. 2016;8(8):832-8.

150. Munyeza CF, Shobo A, Baijnath S, Bratkowska D, Naiker S, Bester LA, et al. Rapid and widespread distribution of doxycycline in rat brain: a mass spectrometric imaging study. Xenobiotica. 2016;46(5):385-92.

151. Shobo A, Bratkowska D, Baijnath S, Naiker S, Somboro AM, Bester LA, et al. Tissue distribution of pretomanid in rat brain via mass spectrometry imaging. Xenobiotica. 2016;46(3):247-52.

152. Shobo A, Bratkowska D, Baijnath S, Naiker S, Bester LA, Singh SD, et al. Visualization of Time-Dependent Distribution of Rifampicin in Rat Brain Using MALDI MSI and Quantitative LCMS/MS. Assay Drug Dev Technol. 2015;13(5):277-84.

153. Lee SC, Dickson DW, Casadevall A. Pathology of cryptococcal meningoencephalitis: analysis of 27 patients with pathogenetic implications. Hum Pathol. 1996;27(8):839-47.

154. Charlier C, Dromer F, Leveque C, Chartier L, Cordoliani YS, Fontanet A, et al. Cryptococcal neuroradiological lesions correlate with severity during cryptococcal meningoencephalitis in HIV-positive patients in the HAART era. PLoS ONE [Electronic Resource]. 2008;3(4):e1950.

155. Lan SH, Chang WN, Lu CH, Lui CC, Chang HW. Cerebral infarction in chronic meningitis: a comparison of tuberculous meningitis and cryptococcal meningitis. Qjm. 2001;94(5):247-53.

156. Chen SF, Lu CH, Lui CC, Huang CR, Chuang YC, Tan TY, et al. Acute/subacute cerebral infarction (ASCI) in HIV-negative adults with cryptococcal meningoencephalitis (CM): a MRI-based follow-up study and a clinical comparison to HIV-negative CM adults without ASCI. BMC Neurology. 2011;11:12.

157. Jarvis JN, Meintjes G, Rebe K, Williams GN, Bicanic T, Williams A, et al. Adjunctive interferon-gamma immunotherapy for the treatment of HIV-associated cryptococcal meningitis: a randomized controlled trial. Aids. 2012;26(9):1105-13.

158. Jarvis JN, Bicanic T, Loyse A, Namarika D, Jackson A, Nussbaum JC, et al. Determinants of mortality in a combined cohort of 501 patients with HIV-associated Cryptococcal meningitis: implications for improving outcomes. Clin Infect Dis. 2014;58(5):736-45.

159. Speed B, Dunt D. Clinical and host differences between infections with the two varieties of Cryptococcus neoformans. Clin Infect Dis. 1995;21(1):28-34; discussion 5-6.

160. CDC. Emergence of Cryptococcus gattii-- Pacific Northwest, 2004-2010. MMWR Morb Mortal Wkly Rep. 2010;59(28):865-8.

161. Chen SC, Slavin MA, Heath CH, Playford EG, Byth K, Marriott D, et al. Clinical manifestations of Cryptococcus gattii infection: determinants of neurological sequelae and death. Clin Infect Dis. 2012;55(6):789-98.

162. Stockamp NW, Thompson GR. Coccidioidomycosis. Infectious Disease Clinics of North America. 2016;30(1):229-46.

163. de Carvalho CA, Allen JN, Zafranis A, Yates AJ. Coccidioidal meningitis complicated by cerebral arteritis and infarction. Human Pathology. 1980;11(3):293-6.

164. Stevens DA, Shatsky SA. Intrathecal amphotericin in the management of coccidioidal meningitis. Semin Respir Infect. 2001;16(4):263-9.

165. Berry CD, Happs EL, Sahrakar K, Stevens DA, Hassid EI, Pappagianis D. A New Method for the Treatment of Chronic Fungal Meningitis: Continuous Infusion into the Cerebrospinal Fluid for Coccidioidal Meningitis. The American Journal of the Medical Sciences. 2009;338(1):79-82.

166. Goldstein EJC, Johnson RH, Einstein HE. Coccidioidal Meningitis. Clinical Infectious Diseases. 2006;42(1):103-7.

167. Garg N, Devi IB, Vajramani GV, Nagarathna S, Sampath S, Chandramouli BA, et al. Central nervous system cladosporiosis: an account of ten culture-proven cases. Neurol India. 2007;55(3):282-8.

168. Dixon DM, Walsh TJ, Merz WG, McGinnis MR. Infections due to Xylohypha bantiana (Cladosporium trichoides). Rev Infect Dis. 1989;11(4):515-25.

169. Filizzola MJ, Martinez F, Rauf SJ. Phaeohyphomycosis of the central nervous system in immunocompetent hosts: report of a case and review of the literature. Int J Infect Dis. 2003;7(4):282-6.

170. Revankar SG, Sutton DA, Rinaldi MG. Primary central nervous system phaeohyphomycosis: a review of 101 cases. Clin Infect Dis. 2004;38(2):206-16.

171. Ashdown BC, Tien RD, Felsberg GJ. Aspergillosis of the brain and paranasal sinuses in immunocompromised patients: CT and MR imaging findings. AJR Am J Roentgenol. 1994;162(1):155-9.

172. Wang RX, Zhang JT, Chen Y, Huang XS, Jia WQ, Yu SY. Cerebral aspergillosis: a retrospective analysis of eight cases. Int J Neurosci. 2017;127(4):339-43.

173. Pongbhaesaj P, Dejthevaporn C, Tunlayadechanont S, Witoonpanich R, Sungkanuparph S, Vibhagool A. Aspergillosis of the central nervous system: a catastrophic opportunistic infection. Southeast Asian J Trop Med Public Health. 2004;35(1):119-25.

174. Spapen H, Spapen J, Taccone FS, Meersseman W, Rello J, Dimopoulos G, et al. Cerebral aspergillosis in adult critically ill patients: a descriptive report of 10 patients from the AspICU cohort. Int J Antimicrob Agents. 2014;43(2):165-9.

175. Hamill RJ. Fungal infections of the central nervous system. In: Anaissie EJ, McGinnis MR, Phaller MA, editors. Clinical Mycology. Second ed: Churchill Livingstone (Elselvier); 2009. p. 591 - 608.

176. Rodriguez-Tudela JL, Berenguer J, Guarro J, Kantarcioglu AS, Horre R, de Hoog GS, et al. Epidemiology and outcome of Scedosporium prolificans infection, a review of 162 cases. Med Mycol. 2009;47(4):359-70.

177. Lamaris GA, Chamilos G, Lewis RE, Safdar A, Raad II, Kontoyiannis DP. Scedosporium Infection in a Tertiary Care Cancer Center: A Review of 25 Cases from 1989–2006. Clinical Infectious Diseases. 2006;43(12):1580-4.

178. Troke P, Aguirrebengoa K, Arteaga C, Ellis D, Heath CH, Lutsar I, et al. Treatment of scedosporiosis with voriconazole: clinical experience with 107 patients. Antimicrob Agents Chemother. 2008;52(5):1743-50.

179. Husain S, Munoz P, Forrest G, Alexander BD, Somani J, Brennan K, et al. Infections due to Scedosporium apiospermum and Scedosporium prolificans in transplant recipients: clinical characteristics and impact of antifungal agent therapy on outcome. Clin Infect Dis. 2005;40(1):89-99.

180. Sanchez-Portocarrero J, Perez-Cecilia E, Corral O, Romero-Vivas J, Picazo JJ. The central nervous system and infection by Candida species. Diagn Microbiol Infect Dis. 2000;37(3):169-79.

181. Voice RA, Bradley SF, Sangeorzan JA, Kauffman CA. Chronic candidal meningitis: an uncommon manifestation of candidiasis. Clin Infect Dis. 1994;19(1):60-6.

182. Casado JL, Quereda C, Oliva J, Navas E, Moreno A, Pintado V, et al. Candidal meningitis in HIV-infected patients: analysis of 14 cases. Clin Infect Dis. 1997;25(3):673-6.

183. Benjamin DK, Jr., Stoll BJ, Fanaroff AA, McDonald SA, Oh W, Higgins RD, et al. Neonatal candidiasis among extremely low birth weight infants: risk factors, mortality rates, and neurodevelopmental outcomes at 18 to 22 months. Pediatrics. 2006;117(1):84-92.

184. Nguyen MH, Yu VL. Meningitis caused by Candida species: an emerging problem in neurosurgical patients. Clin Infect Dis. 1995;21(2):323-7.

185. Schestatsky P, Chedid MF, Amaral OB, Unis G, Oliveira FM, Severo LC. Isolated central nervous system histoplasmosis in immunocompetent hosts: a series of 11 cases. Scand J Infect Dis. 2006;38(1):43-8.

186. Saccente M, McDonnell RW, Baddour LM, Mathis MJ, Bradsher RW. Cerebral histoplasmosis in the azole era: report of four cases and review. South Med J. 2003;96(4):410-6.

187. Wheat LJ, Musial CE, Jenny-Avital E. Diagnosis and management of central nervous system histoplasmosis. Clin Infect Dis. 2005;40(6):844-52.

188. Freifeld A, Proia L, Andes D, Baddour LM, Blair J, Spellberg B, et al. Voriconazole use for endemic fungal infections. Antimicrob Agents Chemother. 2009;53(4):1648-51.

189. Thompson GR, 3rd, Wiederhold NP. Isavuconazole: a comprehensive review of spectrum of activity of a new triazole. Mycopathologia. 2010;170(5):291-313.

190. Black KE, Baden LR. Fungal infections of the CNS: treatment strategies for the immunocompromised patient. CNS Drugs. 2007;21(4):293-318.

191. Gonyea EF. The spectrum of primary blastomycotic meningitis: A review of central nervous system blastomycosis. Annals of Neurology. 1978;3(1):26-39.

192. Chapman SW, Dismukes WE, Proia LA, Bradsher RW, Pappas PG, Threlkeld MG, et al. Clinical practice guidelines for the management of blastomycosis: 2008 update by the Infectious Diseases Society of America. Clin Infect Dis. 2008;46(12):1801-12.

193. Trifilio SM, Bennett CL, Yarnold PR, McKoy JM, Parada J, Mehta J, et al. Breakthrough zygomycosis after voriconazole administration among patients with hematologic malignancies who receive hematopoietic stem-cell transplants or intensive chemotherapy. Bone Marrow Transplant. 2007;39(7):425-9.

194. Bitar D, Van Cauteren D, Lanternier F, Dannaoui E, Che D, Dromer F, et al. Increasing incidence of zygomycosis (mucormycosis), France, 1997-2006. Emerg Infect Dis. 2009;15(9):1395-401.

195. Aristizabal BH, Clemons KV, Cock AM, Restrepo A, Stevens DA. Experimental paracoccidioides brasiliensis infection in mice: influence of the hormonal status of the host on tissue responses. Med Mycol. 2002;40(2):169-78.

196. Elias J, Jr., dos Santos AC, Carlotti CG, Jr., Colli BO, Canheu A, Matias C, et al. Central nervous system paracoccidioidomycosis: diagnosis and treatment. Surg Neurol. 2005;63 Suppl 1:S13-21; discussion S.

197. Queiroz-Telles F, Goldani LZ, Schlamm HT, Goodrich JM, Espinel-Ingroff A, Shikanai-Yasuda MA. An open-label comparative pilot study of oral voriconazole and itraconazole for long-term treatment of paracoccidioidomycosis. Clin Infect Dis. 2007;45(11):1462-9.

198. Surmont I, Vergauwen B, Marcelis L, Verbist L, Verhoef G, Boogaerts M. First report of chronic meningitis caused byTrichosporon beigelii. European Journal of Clinical Microbiology and Infectious Diseases. 1990;9(3):226-9.

199. Watson KC, Kallichurum S. Brain abscess due to Trichosporon cutaneum. J Med Microbiol. 1970;3(1):191-3.

200. Gottfredsson M, Perfect JR. Fungal meningitis. Semin Neurol. 2000;20(3):307-22.

**\*\*Comprehensive review of fungal meningitis.**

201. Pasko MT, Piscitelli SC, Van Slooten AD. Fluconazole: a new triazole antifungal agent. Dicp. 1990;24(9):860-7.

202. Troke PF, Andrews RJ, Pye GW, Richardson K. Fluconazole and Other Azoles: Translation of in Vitro Activity to in Vivo and Clinical Efficacy. Reviews of Infectious Diseases. 1990;12(Supplement\_3):S276-S80.

203. Gubbins PA, EJ. Antifungal therapy. In: Anaissie EM, MR; Pfaller, MA, editor. Clinical Mycology. 2nd ed: Churchill Livingstone, Elsevier; 2009. p. 161 - 96.

204. Girmenia C. New generation azole antifungals in clinical investigation. Expert Opin Investig Drugs. 2009;18(9):1279-95.

205. NationalCenterforBiotechnologyInformation. Isavuconazonium. PubChem Compound Database; CID=6918606: National Center for Biotechnology Information; [cited 2017 November 8]. Available from: <https://pubchem.ncbi.nlm.nih.gov/compound/6918606>

206. Bekersky I, Fielding RM, Dressler DE, Lee JW, Buell DN, Walsh TJ. Plasma protein binding of amphotericin B and pharmacokinetics of bound versus unbound amphotericin B after administration of intravenous liposomal amphotericin B (AmBisome) and amphotericin B deoxycholate. Antimicrobial Agents & Chemotherapy. 2002;46(3):834-40.

207. Hartsel S, Bolard J. Amphotericin B: new life for an old drug. Trends in Pharmacological Sciences. 1996;17(12):445-9.

208. Clemons KV, Espiritu M, Parmar R, Stevens DA. Comparative efficacies of conventional amphotericin b, liposomal amphotericin B (AmBisome), caspofungin, micafungin, and voriconazole alone and in combination against experimental murine central nervous system aspergillosis. Antimicrob Agents Chemother. 2005;49(12):4867-75.

209. Hospenthal DR, Rinaldi MG. Diagnosis and Treatment of Fungal Infections: Springer International Publishing; 2015.

210. P L, U W, A S-H. Isavuconazole and Other Azoles with Respect to Physicochemical and Pharmacokinetic Properties Affecting Tissue Penetration. ECCMID; Vienna2017.