# Suppressive drug interactions between antifungals

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# Suppressive Drug Interactions between Antifungals

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**In this issue of *Chemistry & Biology*, Cokol and colleagues report a systematic study of drug interactions between antifungal compounds. Suppressive drug interactions occur more frequently than previously realized and come in different flavors with interesting implications.**

When two drugs are combined, they may interact synergistically or antagonistically; for synergistic interactions, the combined drug effect on growth is stronger than expected, while for antagonistic interactions it is weaker (Figures 1A–1C) (Greco et al., 1995; Loewe, 1928). Suppressive interactions are hyper-antagonistic cases in which the addition of one drug on top of another actually increases growth—a surprising effect, given that both drugs alone inhibit growth (Figure 1D). Drug interactions are analogous to genetic epistasis in which the combined effect of genetic perturbations is characterized by its deviation from additivity (Poelwijk et al., 2007). Most research has focused on synergistic drugs, because they allow the use of lower drug dosages, which can facilitate treatments. In contrast, antagonistic and suppressive drug interactions have received less attention, although they have been known for a long time. Over 140 years ago, Fraser measured a two-drug response surface in rabbits and observed that the effect of *Physostigma venenosum* extract was suppressed by atropine (Fraser, 1871)—rabbits given just the right dose of both drugs were hopping around happily, whereas the same dose of physostigma alone killed the animals within minutes.

A possible reason for the lack of attention to suppressive drug combinations is that they can evidently impair treatment efficiency. Interestingly though, recent work on antibiotic combinations has shown that suppressive drug combinations can lead to selection against drug resistance (Chait et al., 2007). Thus, suppressive drug interactions may have important implications for the long-term success of treatments. Beyond medical applications, drug combinations are generally an important means of controlled cellular perturbation. Compared to genetic perturbations, drugs have the advantage that their dose can be altered continuously, which may facilitate the elucidation of complex relationships between cellular functions. However, few systematic studies characterizing pairwise interactions between large sets of drugs exist, and in particular the prevalence of suppressive interactions in these networks has not been systematically mapped.

In this issue of *Chemistry & Biology*, Cokol et al. (2014) investigated the pervasiveness of suppression between many antifungal drug pairs in *Saccharomyces cerevisiae*. They found that 17% of 175 tested drug pairs showed suppressive interactions. This high frequency of suppressive interactions is a conservative estimate of the prevalence of suppression between antifungals, because the drug pairs in this data set were originally chosen in an attempt to identify synergistic interactions (Cokol et al., 2011). By assaying another 40 drug combinations (consisting of 10 unbiased drugs tested against 4 drugs with a tendency to suppress or be suppressed), the authors found an even higher fraction of suppressive drug interactions of 38%. Overall, these observations suggest that suppression between antifungals occurs quite frequently.

Suppressive interactions typically have a direction, i.e., one drug suppresses the effect of the other, but not the other way around (Figure 1D). Interestingly, apart from such directed suppression, Cokol et al. (2014) observed several reciprocally suppressive interactions in which drug A suppressed drug B and vice versa (Figures 1E and 1F). This special type of suppression is interesting, because theoretical arguments suggest that a reciprocally suppressive drug combination as in Figure 1E can lead to selection against mutants that are resistant to either drug alone (Chait et al., 2007), leaving only mutations that confer resistance to both drugs simultaneously as a viable path for the emergence of drug resistance. Such mutations are expected to be rare, and reciprocally suppressive drugs could thus slow down the evolution of drug resistance: at least as long as the drug interaction itself is not altered by mutations. Although most examples of reciprocal suppression reported by Cokol et al. (2014) are more similar to the case in Figure 1F, it will be interesting to test these newly found reciprocally suppressive drug pairs for their ability to select against drug resistance.

Another observation made by the authors is that the antifungal compound Bromopyruvate (Bro) often suppressed other drugs while another (Staurosporine [Sta]) was more often suppressed (Cokol et al., 2014). This extends previous findings that certain antifungals often show synergistic interactions with other drugs in yeast (Cokol et al., 2011) and that aminoglycoside antibiotics often interact synergistically with other antibiotics in bacteria (Yeh et al., 2006). Overall, a general property of drug interaction networks is emerging; certain drugs form network hubs that are highly enriched for the same type of interaction with other drugs.

Intriguingly, this observation suggests that there may be general underlying causes of these drug interactions; e.g., a drug could trigger the upregulation of a multidrug efflux pump protecting cells from many other drugs, which would be

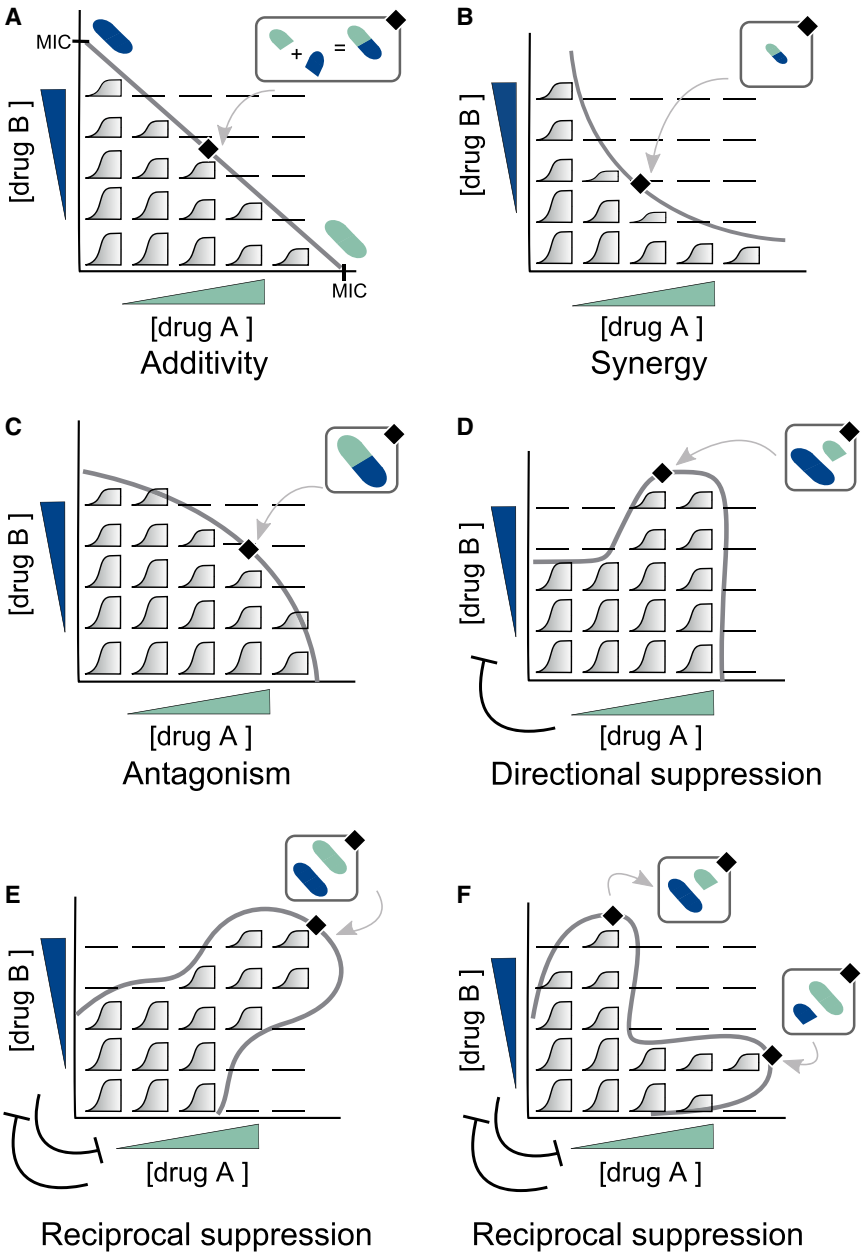


**Figure 1. Drug Interactions**

Shown are schematic response surfaces representing growth of a microbial culture in 5×5 concentration matrices. The growth response to the drugs is depicted as the area under the growth curve. The dose of both drugs that is needed to reach the black diamond on the minimal inhibitory concentration (MIC) line (gray) is indicated by pills of corresponding size.

(A) Additivity: adding a half-MIC dose of each drug A and drug B leads to the same growth inhibition as a full-MIC dose of either drug A or drug B.

(B) Synergy: the drugs together inhibit growth more strongly than expected from additivity.

(C) Antagonism: the combined effect of the drugs is weaker than expected from additivity.

(D) Directional suppression: a higher dose of drug B is needed to reach the MIC line in presence of drug A than in its absence.

(E and F) Reciprocal suppression: both drugs suppress each other.

reflected by suppressive interactions with these drugs. This is currently speculation though, and, generally, little is known about the causes of drug interactions. To gain insight into the causes of a selected suppressive interaction (between Bro and Sta) Cokol et al. (2014) used haploinsufficiency profiling and homozygous deletion profiling. Although these chemogenomic analyses yielded valuable insights into the individual drug effects (e.g., the effect of Bro was modulated in *acs2*Δ/*ACS2* and *erg10*Δ/*ERG10* mutants, which both affect the same metabolic pathway), they could not reveal the mechanism of suppression between the two drugs. Their work further adds a note of caution to drug interaction studies; a small pH change in the growth medium, due to the addition of certain drugs, was found to suppress certain other drugs.

The difficulty in identifying the causes of drug interactions may be due to complex effects on multiple cellular functions. An example from bacteria where the interaction mechanism has been characterized is the suppression between translation and DNA synthesis inhibitors. These were shown to be caused by a nonoptimal regulation of ribosome production under DNA synthesis inhibitors, which is partly corrected by the translation inhibitor (Bollenbach et al., 2009). It will be an interesting direction for future research to elucidate the causes of the newly observed suppressive interactions between antifungals (Cokol et al., 2014).

## REFERENCES

Bollenbach, T., Quan, S., Chait, R., and Kishony, R. (2009). Cell *139*, 707–718.

Chait, R., Craney, A., and Kishony, R. (2007). Nature *446*, 668–671.

Cokol, M., Chua, H.N., Tasan, M., Mutlu, B., Weinstein, Z.B., Suzuki, Y., Nergiz, M.E., Costanzo, M., Baryshnikova, A., Giaever, G., et al. (2011). Mol. Syst. Biol. *7*, 544.

Cokol, M., Weinstein, Z.B., Yilancioglu, K., Tasan, M., Doak, A., Cansever, D., Mutlu, B., Li, S., Rodriguez-Esteban, R., Akhmedov, M., et al. (2014). Chem. Biol. *21*, this issue, 541–551.

Fraser, T.R. (1871). Proc Roy Soc Edin. *7*, 506–511.

Greco, W.R., Bravo, G., and Parsons, J.C. (1995). Pharmacol. Rev. *47*, 331–385.

Loewe, S. (1928). Ergeb Physiol Biol Chem Exp Pharmakol *27*, 47–187.

Poelwijk, F.J., Kiviet, D.J., Weinreich, D.M., and Tans, S.J. (2007). Nature *445*, 383–386.

Yeh, P., Tschumi, A.I., and Kishony, R. (2006). Nat. Genet. *38*, 489–494.

 