# *Ex vivo* permeability study of poorly soluble drugs across gastrointestinal membranes: acceptor compartment media composition

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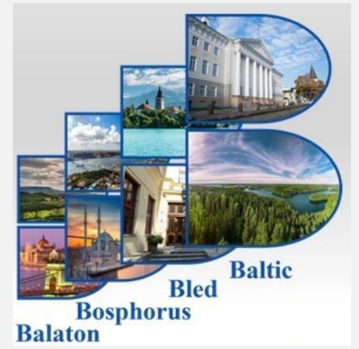
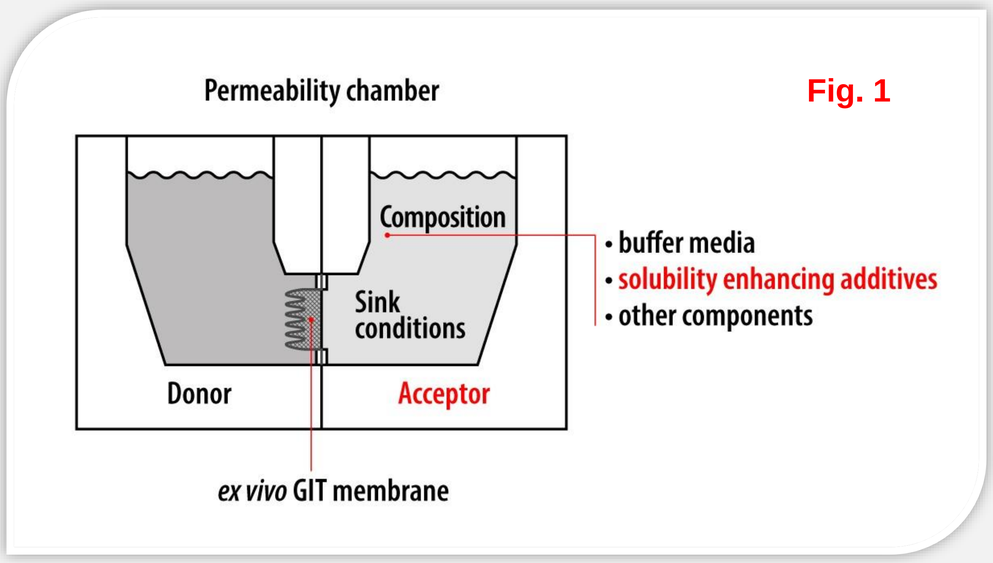



## Introduction

- In *ex vivo* drug permeability studies, establishing sink conditions for permeated poorly soluble drugs in the relatively low and/or limited media volumes of chamber's acceptor compartment is crucial (**Fig.1**).
- Adding solubility-enhancing agents to the acceptor media addresses this challenge and reduces non-specific binding of drug molecules to the walls of the permeability chamber and the membrane itself.
- **This study aimed to** investigate and critically evaluate compositions of the acceptor compartment media used in *ex vivo* permeability studies for poorly soluble drugs, in relation to their physicochemical, biopharmaceutical (**Fig. 2 and 3**), and pharmacokinetic properties.




## Methods

- A **literature search** was conducted, focusing on contributions from leading pharmacy schools and research groups.

## Results

- Most commonly reported *ex vivo* acceptor media were KBR, HBSS, and PBS (**Table 1**); media components and compositions varied across laboratories and studies.
- Only 9 studies reported the use of solubility-enhancing additives in acceptor media for *ex vivo* studies (**Table 1**).
- BSA was used in acceptor media where poorly soluble drugs exhibited high plasma protein binding or binding to tissue proteins (**Fig. 2 & 3, Table 1**).
- SLS (HLB 40), was added in a study with the lipophilic drug exemestane (**Fig. 2 & 3, Table 1**).
- Poloxamer 407 (HLB 22) established acceptor compartment sink conditions in studies with lipophilic dapivirine (**Fig. 2 & 3, Table 1**).
- Tween 80 (HLB 15) was applied in studies with lipophilic fenretinide and docetaxel, as well as the hydrophilic drug cisplatin (**Fig. 2 & 3, Table 1**).
- Foaming due to continuous oxygenation *in ex vivo* systems, was reported for BSA and may also occur with surfactants.
- Anti-foaming agents helped resolve foaming issue, although their impact on permeability study results was not discussed.
- The use of surfactants and BSA impacts the analysis of drug concentrations, requiring additional purification steps.
- *In vitro* permeability studies have reported also the use of the following additives to establish sink conditions: TPGS, caprylocaproyl polyoxyl-8 glycerides, β-cyclodextrin, and sodium taurocholate (**Table 3**).

**Table 1**. Acceptor compartment media compositions with solubility enhancing additives reported in published *ex vivo* permeability studies with poorly soluble drugs

| Solubility enhancing additives | Other additives | Name (reported) | Composition, mM | | | | | | | | | | | | pH | Oxygenation ($O_2$/$CO_2$, 95%/5%) | Reported osmolarity (mOs/kg) | Tested drug | GIT membrane origin (species) | Tissue |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | NaCl | KCl | $CaCl_2$ | $CaSO_4$ | $MgCl_2$ | $MgSO_4$ | $NaH_2PO_4$ | $Na_2HPO_4$ | $K_2HPO_4$ | $KH_2PO_4$ | $NaHCO_3$ | Glucose | | | | | | |
| **0.1% BSA** | 25 mM HEPES | KRB | NR | NR | 2.5 | NR | NR | NR | NR | NR | NR | NR | 15 | 10 | 7.4 | Yes | NR | **Digoxin** | Human | Ileum, colon |
| **1% BSA** | 10 mM HEPES | KRB | NR | NR | 1 | NR | NR | NR | NR | NR | NR | NR | 25 | NR | 7.4 | Yes | NR | **Ibuprofen** | Human | Duodenum, jejunum, ileum, colon |
| **0.2% Poloxamer 407** | | HBSS | 140 | 5 | 1 | NR | 0.5 | 0.4 | NR | 0.3 | NR | 0.4 | 4 | 6 | 7.3 | NR | 287 | **Dapivirine** | Pig | Rectal mucosa |
| **0.0084% Tween 80** | | PBS | NR | NR | NR | NR | NR | NR | NR | NR | NR | NR | NR | NR | 7.4 | NR | NR | **Fenretinide** | Pig | Buccal mucosa |
| **0.1% Tween 80** | | PBS | NR | NR | NR | NR | NR | NR | NR | NR | NR | NR | NR | NR | 7.4 | NR | NR | **Docetaxel, cisplatin** | Pig | Esophageal mucosa |
| **0.5% SLS** | | PBS | 95.8 | NR | NR | NR | NR | NR | NR | 16.8 | NR | 1.4 | NR | NR | 7.4 | NR | 290 | **Exemestane** | Pig | Intestinal tissue |
| **0.6 % BSA** | | PBS/Receptor fluid | NR | NR | NR | NR | NR | NR | 100 | NR | NR | NR | NR | NR | 7.4 | NR | 290 | **Diazepam** | Pig | Buccal mucosa |
| **4.5 % BSA** | Antifoam B 0.1 % | HBSS | NR | NR | NR | NR | NR | NR | NR | NR | NR | NR | NR | NR | 7.4 | Yes | NR | **Aprepitant** | Rat | Duodenum, jejunum, ileum |

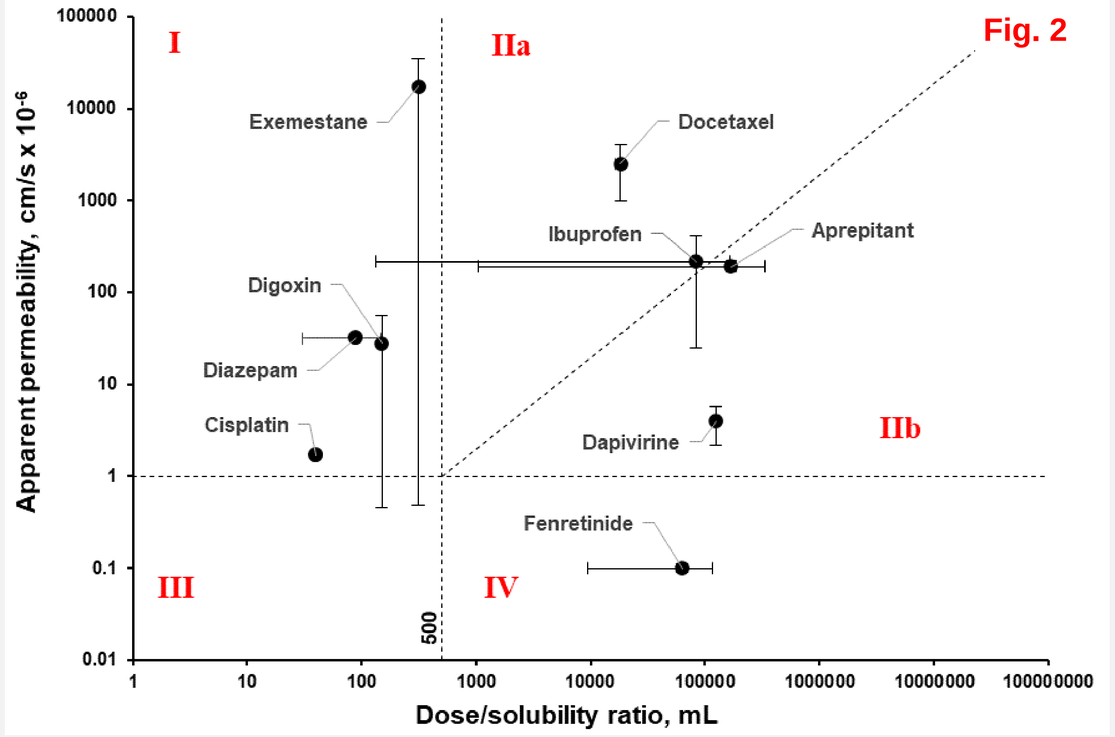



**Table 2**. Acceptor compartment media compositions reported in some cell culture-based permeability studies with poorly soluble drugs

| Solubility enhancing additive | Cell line | Acceptor media | pH | Tested drug |
|---|---|---|---|---|
| **4% BSA** | Caco-2 cell monolayer | HBSS with/without 10 mM morpholinopropanesulfonic acid | 7.4 | **Ritonavir; ABT-102** |
| **0.2% TPGS** | Caco-2 cell monolayer | HBSS with 10 mM HEPES and 25 mM glucose | 7.4 | **Itraconazole** |
| **4% BSA or 0.5% TPGS or 1% DMSO** | Different | KBR | 7.0; 7.4 | **Different** |

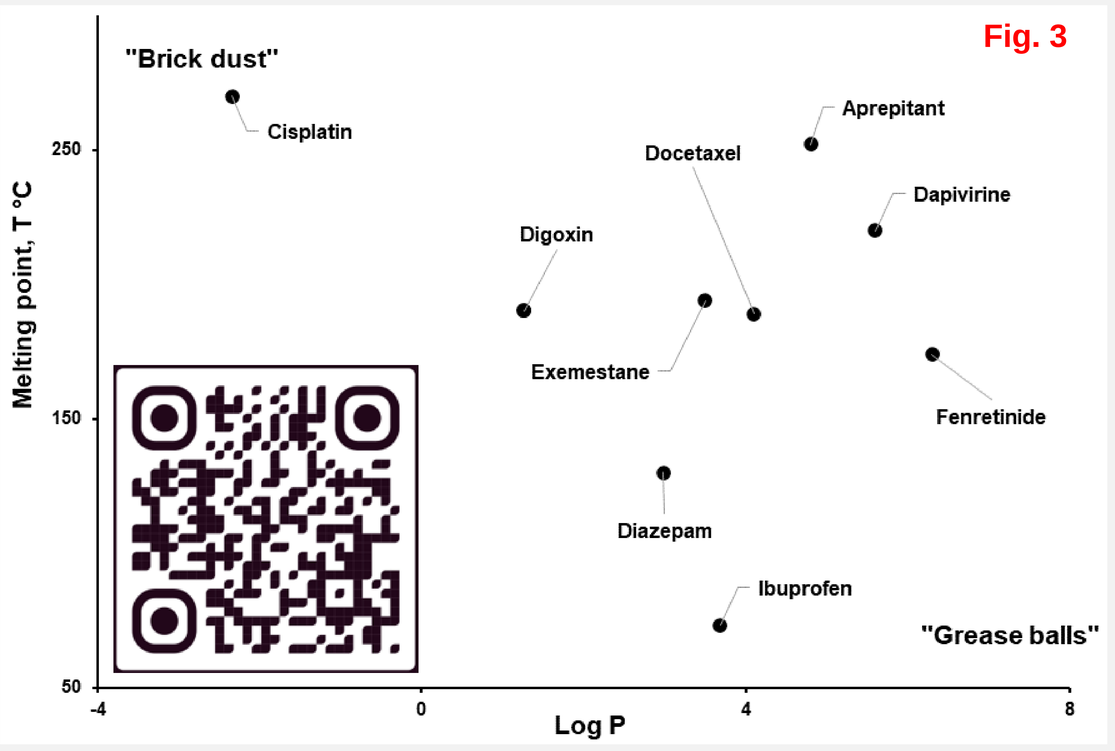



**Table 3**. Acceptor compartment media compositions reported in some *in vitro* artificial membrane-based permeability studies with poorly soluble drugs

| Solubility enhancing additive | Permeable membrane | Acceptor media | pH | Tested drug |
|---|---|---|---|---|
| **1-1.5% SLS** | PAMPA; PermeaLoop™, Microdialysis-dissolution/permeation system | 10 mM HEPES-buffered HBSS; Simulated gastric fluid; PBS | 7.4; 6.5 | **Naproxen; posaconazole, efavirenz; celecoxib** |
| **3-4% BSA** | Double-Sink PAMPA; PermeaPad® | PBS (290 mOsmol/kg) | 7.4 | **Cinnarizin** |
| **0.6% Labrasol®** | Hydrophilic cellulose hydrate membrane | Purified water with NaCl | | **Fenofibrate** |
| **0.2-0.5% TPGS** | PermeaLoop™ | PBS; HBSS | 6.5; 7.35; 7.4 | **ABT-869; dipyridamole, fosamprenavir; itraconazole** |
| **50 mM sodium taurocholate** | Dialysis membrane with a molecular weight cutoff of >1000 Da | 2 mM Tris maleate, 1.4 mM $CaCl_2$, 150 mM NaCl | 6.5 | **Saquinavir** |
| **1% Tween 80** | PermeaLoop™ | PBS | 6.5 | **Celecoxib** |
| **10% Hydroxypropyl-β-cyclodextrin** | PermeaLoop™; PermeaPad® Plate | FaSSIF; HCl solution; PBS | 6.5;1.5 | **Posaconazole; itraconazole** |

## Conclusions

- Despite the clear necessity, the addition of solubility-enhancing additives to increase sink conditions was rarely reported in literature.
- The addition of solubility-enhancing additives may influence drug permeation across *ex vivo* membranes in the serosal-to-mucosal direction. Although not previously reported, this potential effect warrants consideration in future studies.

Sitovs, A.; Mohylyuk, V. "Ex Vivo Permeability Study of Poorly Soluble Drugs across Gastrointestinal Membranes: Acceptor Compartment Media Composition." Poster. 10th BBB Conference on Pharmaceutical Sciences "Today´s science, tomorrow´s healthcare", Tartu, Estonia, September 13, 2024.