E-companion: A Novel Adaptive Testing Scheme for Multi-disease Testing

A. Summary of Notation

Table A.1 Notation for the adaptive testing design problem.

Sets and Vectors:	
$[a]=\{1,2,\cdots,a\}$	Set of integers from 1 to a ($a\in Z^+$)
$\mathcal{N}=[n]$	Set of diseases, with cardinality n
$\mathcal{S}$	Set of diseases targeted by assay $\mathcal{S}$, with cardinality s ($\mathcal{S}\subseteq\mathcal{N}$)
$\mathcal{S}(r)$	Set of all ordered r-combinations of set $\mathcal{S}$ ($r\in[s],\mathcal{S}\subseteq\mathcal{N}$)
$\mathcal{P}^{\texttt{cml}}(\mathcal{N})=\cup_{r\in[n]}\mathcal{N}(r)$	Cumulative assay set of all 2^n-1 nonempty subsets (assays) of set $\mathcal{N}$
$\mathcal{I}(\cdot)$	Index set of a given collection of sets
$\mathcal{I}^{\texttt{cml}}(\mathcal{S})=\cup_{r\in[s]}\mathcal{I}(\mathcal{S}(r))$	Cumulative index set of set $\mathcal{S}$ ($\mathcal{S}\subseteq\mathcal{N}$)
$\{0\}\cup\mathcal{I}^{\texttt{cml}}(\mathcal{N})$	Set of 2^n disease category indices
$\boldsymbol{\mathcal{S}}^{n\text{-plex}}=(\mathcal{N})$	Single-assay portfolio, consisting of one n-plex
$\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}=(\{k\})_{k\in[n]}$	All-singleton portfolio, consisting of n singletons
Parameters:	
$\boldsymbol{p}=\big(p_0,(p_c)_{c\in\mathcal{I}^{\texttt{cml}}(\mathcal{N})}\big)$	Joint prevalence vector
$\boldsymbol{p}(\mathcal{S})=\big(p_0(\mathcal{S}),(p_c(\mathcal{S}))_{c\in\mathcal{I}^{\texttt{cml}}(\mathcal{S})}\big)$	Joint prevalence vector for assay $\mathcal{S}$ ($\mathcal{S}\subseteq\mathcal{N}$)
π_i, $\pi(\mathcal{S})$	Marginal prevalence of disease i ($i\in\mathcal{N}$), prevalence of assay $\mathcal{S}$ ($\mathcal{S}\subseteq\mathcal{N}$)
$\boldsymbol{\pi}$	Marginal prevalence vector for all diseases in set $\mathcal{N}$
$\boldsymbol{p}\in\mathcal{P}(\boldsymbol{\pi})$	Set of joint prevalence vectors consistent with the marginal prevalence vector $\boldsymbol{\pi}$
$\underline{p}(\mathcal{S}),\underline{p}_{\min}(\boldsymbol{\pi})$	Assay-specific ($\mathcal{S}\subseteq\mathcal{N}$) and universal adaptive pooling thresholds
$\underline{p}^{\texttt{Dorf}}=1-e^{-e^{-1}}\approx 0.308$	Dorfman pooling threshold
c_f, c_v	Fixed cost per test, variable cost per disease targeted by the assay
Decisions:	
Main:	
$\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}=(\mathcal{S}_k)_{k\in[q]}$	Assay portfolio (q-partition of set $\mathcal{N}$ for some $q\in[n]$), as a vector of sets (assays)
$\boldsymbol{t}$	Assay pool size vector
Auxiliary:	
$\boldsymbol{w}(\boldsymbol{t})$	Disease pool size vector, where $w_i(\boldsymbol{t})=t_k$ if $i\in\mathcal{S}_k$ ($i\in\mathcal{N}$)
$\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$, $\mathcal{K}_{\texttt{ind}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$	Index set of pooled assays, index set of individually tested assays
$C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$	Adaptive testing cost (i.e., expected testing cost per subject)
For Dorfman Testing:	
$C^{\texttt{Dorf}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$	Testing cost (i.e., expected testing cost per subject) for Dorfman testing
$\boldsymbol{t}^{\texttt{Dorf}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$	Optimal pool size vector for Dorfman testing when $\pi(\mathcal{N})<\underline{p}^{\texttt{Dorf}}$
$\bar{\boldsymbol{t}}^{\texttt{Dorf}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$	Finite local maximum pool size vector for Dorfman cost function when $\pi(\mathcal{N})<\underline{p}^{\texttt{Dorf}}$
Events & Random variables:	
$T_i^+(\mathcal{S},t)$	Event that the test outcome for disease i is positive when assay $\mathcal{S}$ is tested with a random pool of size t ($i\in\mathcal{S}$) (with complement $T_i^-(\mathcal{S},t)$)
$\mathcal{N}_{pool}^+(\mathcal{S},t)=\{i\in\mathcal{S}:T_i^+(\mathcal{S},t)\}$	Pool-level retesting set (with complement $\mathcal{N}_{pool}^-(\mathcal{S},t)$)
$D_{pool}(\mathcal{S},t)$	Cardinality of a random pool-level retesting set, i.e., $\mid N_{pool}^+(\mathcal{S},t)\mid$
$D_{\texttt{subject}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$	Retesting set cardinality for a random subject
Other:	
Superscript $*$	Optimal adaptive testing design, i.e., $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}*},\boldsymbol{t}^*$
Superscripts ($c_v=0$), ($c_f=0$)	Special cost structures: $c_v=0$ and $c_f=0$
Superscripts nc, indep, nested	Special prevalence structures: no-coinfection, independent, and nested (Definition 1)
Subscript $-k$	All indices except k, e.g., $\boldsymbol{t}_{-k}=(t_\ell)_{\ell\in[q]\setminus\{k\}}$
$W_0(\cdot),W_{-1}(\cdot)$	The principal and secondary branches of the Lambert-W function
FOC	First-order condition wrt pool size ($\frac{\partial C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},t_k,\boldsymbol{t}_{-k})}{\partial t_k}=0,k\in[q]$)

We also define $\gamma_i:=1-\pi_i,\ i\in\mathcal{N};\ \gamma(\mathcal{S}):=p_0(\mathcal{S})$ and $\alpha(\mathcal{S}):=\Big(\ln(\frac{1}{(1-\pi(\mathcal{S}))})\Big)^{1/2},\ \mathcal{S}\subseteq\mathcal{N}.$

B. Proofs

We first provide the supporting results from the literature, followed by the proofs for the results in the paper. Commonly used notation is provided in Appendix A; other less common notation is introduced as needed. We use the abbreviations, with respect to (wrt), without loss of generality (wlog), respectively (resp.), and iff (if and only if).

B.1. Supporting Results from the Literature

Proposition B.1 (Dorfman properties) ***(Implied by Lemmas 1, 2, 8, Theorem 1, and Corollary 4 of Aprahamian et al. (2020)).*** *Consider Dorfman testing for a single disease with prevalence π under perfect test sensitivity and specificity. If $\pi < \underline{p}^{Dorf} := 1 - e^{-e^{-1}} \approx 0.308$, i.e., Dorfman pooling threshold, then the following results hold:*

1. *The expected number of tests per subject function, given by $\frac{1}{t} + 1 - (1-\pi)^t$ at pool size t, has exactly two pool size solutions that satisfy the FOC:*
$$t^{Dorf}(\pi) := \frac{2}{\ln(1-\pi)} \cdot W_0\Big(-\frac{1}{2}\big(\ln(\frac{1}{1-\pi})\big)^{1/2}\Big),\ \bar{t}^{Dorf}(\pi) := \frac{2}{\ln(1-\pi)} \cdot W_{-1}\Big(-\frac{1}{2}\big(\ln(\frac{1}{1-\pi})\big)^{1/2}\Big),$$
where $t^{Dorf}(\pi)$ and $\bar{t}^{Dorf}(\pi)$ resp. correspond to the global minimum and the finite local maximum of the Dorfman cost function, where $t^{Dorf}(\pi) < \bar{t}^{Dorf}(\pi)$.
2. *Pooling reduces the expected number of tests over individual testing: $\frac{1}{t^{Dorf}(\pi)} + 1 - (1-\pi)^{t^{Dorf}(\pi)} < 1$.*
3. *$t^{Dorf}(\pi)$ is decreasing in $\pi \in (0, \underline{p}^{Dorf})$.*

Property B.1 (Multi-disease Dorfman pool size) ***(From Bish et al. (2024)).*** *For any assay $\mathcal{S} \subseteq \mathcal{N}$, the optimal pool size follows by Proposition B.1 by letting $\pi = \pi(\mathcal{S})$.*

We use the notation $t^{\text{Dorf}}(\pi(\mathcal{S}))$ and $t^{\text{Dorf}}(\mathcal{S})$, also $\bar{t}^{\text{Dorf}}(\pi(\mathcal{S}))$ and $\bar{t}^{\text{Dorf}}(\mathcal{S})$, interchangeably.

Property B.2 (Dorfman and Lambert function properties).

1. ***(Based on properties in Aprahamian et al. (2020)).*** *$\bar{t}^{Dorf}(\pi)$ is decreasing in $\pi \in (0, \underline{p}^{Dorf})$.*
2. ***(From Corless et al. (1996)).*** *Both solutions to the Lambert function, $W_0(x)$ and $W_{-1}(x)$, exist only if $x \in (\frac{-1}{e}, 0)$; in this range, $W_0(x) \in (-1, 0)$ and is increasing in x, while $W_{-1}(x) \in (-\infty, -1)$ and is decreasing in x.*

Corollary B.1 (Dorfman pool size relationships). *Let $\mathcal{S} = \{i_1, i_2, \cdots i_s\} \subseteq \mathcal{N}$, with cardinality $s \geq 1$, and diseases indexed following a nonincreasing order of marginal prevalences, i.e., $\pi_{i_1} \geq \pi_{i_2} \geq \cdots \geq \pi_{i_s}$. When $\pi(\mathcal{N}) < \underline{p}^{Dorf}$, Dorfman pool sizes satisfy the following relationships:*
$$t^{Dorf}(\mathcal{N}) \leq t^{Dorf}(\mathcal{S}) \leq t^{Dorf}(\{i_1\}) \leq t^{Dorf}(\{i_2\}) \leq \cdots \leq t^{Dorf}(\{i_s\}),$$
$$\bar{t}^{Dorf}(\mathcal{N}) \leq \bar{t}^{Dorf}(\mathcal{S}) \leq \bar{t}^{Dorf}(\{i_1\}) \leq \bar{t}^{Dorf}(\{i_2\}) \leq \cdots \leq \bar{t}^{Dorf}(\{i_s\}).$$

Proof The result follows from Proposition B.1 and Property B.2, because for any set $\mathcal{S} \subseteq \mathcal{N} : \mathcal{S} \neq \emptyset$, $\pi(\mathcal{N}) \geq \pi(\mathcal{S}) \geq \pi_{i_1} \geq \pi_{i_2} \geq \cdots \geq \pi_{i_s}$. □

B.2. Proof for §2.1

Proof of Lemma 1 For a random subject, let D_j^+ denote the event that the subject is positive for disease j, and E_c the event that the subject belongs to disease category c. Then $\pi(\mathcal{S}) = \mathbb{P}\left(\bigcup_{j \in \mathcal{S}} D_j^+\right)$. Under the nested structure, only the following disjoint disease categories (indices c) have nonzero probability: the no-disease category $c = 0$, singleton categories $c \in \{1, \dots, n\}$, and nested categories $c = 1..m$ for $m \in \{2, \dots, n\}$; disease j is present in any category containing j. Letting $\mathcal{N}_j := \bigcup_{m=\max\{2,j\}}^{n} E_{1..m}$ denote the union of nested categories containing disease $j \in \mathcal{N}$,
$$D_j^+ = E_j \cup \mathcal{N}_j,\ j \in \mathcal{N}. \tag{9}$$
Since $i_{\texttt{min}}(\mathcal{S}) = \min\{i : i \in \mathcal{S}\}$ by definition, we have $i_{\texttt{min}}(\mathcal{S}) \leq j,\ \forall j \in \mathcal{S}$. This implies $\mathcal{N}_j \subseteq \mathcal{N}_{i_{\texttt{min}}(\mathcal{S})},\ \forall j \in \mathcal{S}$ (since any nested category $c = 1..m$ containing j also contains $i_{\texttt{min}}(\mathcal{S})$). Thus:
$$\bigcup_{j \in \mathcal{S}} D_j^+ = \left(\bigcup_{j \in \mathcal{S}} \mathcal{N}_j\right) \cup \left(\bigcup_{j \in \mathcal{S}} E_j\right)$$

$$
\begin{aligned}
&= \mathcal{N}_{i_{\texttt{min}}(\mathcal{S})} \;\cup\; E_{i_{\texttt{min}}(\mathcal{S})} \;\cup \bigcup_{j \in \mathcal{S}\setminus\{i_{\texttt{min}}(\mathcal{S})\}} E_j \\
&= D^+_{i_{\texttt{min}}(\mathcal{S})} \;\cup \bigcup_{j \in \mathcal{S}\setminus\{i_{\texttt{min}}(\mathcal{S})\}} E_j \quad \text{(by (9))},
\end{aligned}
$$

where $D^+_{i_{\texttt{min}}(\mathcal{S})}$ and the singleton category events E_j, $j \neq i_{\texttt{min}}(\mathcal{S})$, are all disjoint. By Definition 1,

$$
\pi(\mathcal{S}) = \mathbb{P}(D^+_{i_{\texttt{min}}(\mathcal{S})}) + \sum_{j \in \mathcal{S}\setminus\{i_{\texttt{min}}(\mathcal{S})\}} p_j = \mathbb{P}(D^+_{i_{\texttt{min}}(\mathcal{S})}) + (s-1)\epsilon = \pi_{i_{\texttt{min}}(\mathcal{S})} + (s-1)\epsilon. \qquad \square
$$

B.3. Proofs for §3.1

B.3.1. Supporting Results For assay $\mathcal{S} \subseteq \mathcal{N}$, the assay's joint prevalence vector consists of:

$$
p_c(\mathcal{S}) := p_c \;+ \sum_{c' \in \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\mathcal{S})} p_{c \oplus c'}, \; \forall c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S}), \qquad p_0(\mathcal{S}) := 1 - \pi(\mathcal{S}) = 1 - \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S})} p_c(\mathcal{S}), \tag{10}
$$

with the special case, $p_0(\mathcal{N}) = 1 - \pi(\mathcal{N}) = p_0$, and $p_c(\mathcal{N}) = p_c$, $\forall c \in \mathcal{I}^{\texttt{cml}}(\mathcal{N})$.

Lemma B.1 (Link between assay and disease prevalences). *For any $\mathcal{S} \subseteq \mathcal{N}$:*

$$
\pi_i = p_i(\mathcal{S}) + \sum_{c \in \mathcal{I}^{cml}(\mathcal{S}\setminus\{i\})} p_{i\oplus c}(\mathcal{S}), \qquad \forall i \in \mathcal{S}.
$$

Proof Consider any $\mathcal{S} \subseteq \mathcal{N}$ and $i \in \mathcal{S}$. Starting with the RHS, we derive the LHS. By (10),

$$
p_i(\mathcal{S}) \overset{(10)}{=} p_i \;+ \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\mathcal{S})} p_{i\oplus c} \quad \text{and} \quad p_{i\oplus c}(\mathcal{S}) \overset{(10)}{=} p_{i\oplus c} \;+ \sum_{c' \in \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\mathcal{S})} p_{i\oplus c\oplus c'}, \;\; \forall i \oplus c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S}), \text{ leading to:}
$$

$$
\begin{aligned}
p_i(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S}\setminus\{i\})} p_{i\oplus c}(\mathcal{S}) &\overset{(10)}{=} \left[p_i \;+ \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\mathcal{S})} p_{i\oplus c} \right] + \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S}\setminus\{i\})} \left[p_{i\oplus c} \;+ \sum_{c' \in \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\mathcal{S})} p_{i\oplus c\oplus c'} \right] \\
&= p_i \;+ \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\mathcal{S})} p_{i\oplus c} \;+ \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S}\setminus\{i\})} p_{i\oplus c} \;+ \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S}\setminus\{i\})} \; \sum_{c' \in \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\mathcal{S})} p_{i\oplus c\oplus c'} = p_i \;+ \sum_{c \in \mathcal{C}} p_{i\oplus c} \\
&= p_i \;+ \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\{i\})} p_{i\oplus c} \;=\; \pi_i \quad \text{(by (1))},
\end{aligned}
$$

where $\mathcal{C} := \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\mathcal{S}) \bigcup \mathcal{I}^{\texttt{cml}}(\mathcal{S}\setminus\{i\}) \bigcup \left[\mathcal{I}^{\texttt{cml}}(\mathcal{S}\setminus\{i\}) \oplus \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\mathcal{S})\right]$, hence $\mathcal{C} = \mathcal{I}^{\texttt{cml}}(\mathcal{N}\setminus\{i\})$. $\square$

Let $\boldsymbol{X}(\mathcal{S},t) = \big(X_0(\mathcal{S},t), (X_c(\mathcal{S},t))_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S})}\big)$ denote the number of subjects with no diseases from set $\mathcal{S}$ ($X_0(\mathcal{S},t)$) and with only the diseases in category c ($X_c(\mathcal{S},t)$), resp., within a random pool of size $t \geq 2$ for assay $\mathcal{S} \subseteq \mathcal{N}$, where index set $\mathcal{I}^{\texttt{cml}}(\mathcal{S})$ contains $2^s - 1$ disease category indices (except for $c = 0$). Observe that $\boldsymbol{X}(\mathcal{S},t) \sim \text{Multinomial}(t, \boldsymbol{p}(\mathcal{S}))$.

Lemma B.2 (Multinomial probabilities). *For any $\mathcal{C}' = \{c_1, c_2, \cdots, c_\ell\} \subseteq \mathcal{I}^{cml}(\mathcal{S})$, $1 \leq \ell \leq 2^s - 1$:*

$$
\mathbb{P}\Big(X_c(\mathcal{S},t) = 0, \; \forall c \in \mathcal{I}^{cml}(\mathcal{S}) \setminus \mathcal{C}'\Big) = \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{C}'} p_c(\mathcal{S})\Big)^t, \;\; \textit{and}
$$

$$
\begin{aligned}
&\mathbb{P}\Big(X_c(\mathcal{S},t) \geq 1, \; \forall c \in \mathcal{C}', \; X_g(\mathcal{S},t) = 0, \; \forall g \in \mathcal{I}^{cml}(\mathcal{S}) \setminus \mathcal{C}'\Big) \\
&\quad = (-1)^\ell \cdot (p_0(\mathcal{S}))^t + \sum_{r=1}^{\ell} (-1)^{\ell - r} \cdot \left[\sum_{\mathcal{C} \in \mathcal{C}'(r)} \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{C}} p_c(\mathcal{S})\Big)^t \right].
\end{aligned}
$$

Proof The first expression readily follows from the multinomial distribution, because the event $\{X_c(\mathcal{S},t)=0,\ \forall c\in\mathcal{I}^{\texttt{cml}}(\mathcal{S})\setminus\mathcal{C}'\}$ implies that all t subjects belong to either category 0 or one of the categories in $\mathcal{C}'$, which happens with probability $p_0(\mathcal{S})+\sum_{c\in\mathcal{C}'}p_c(\mathcal{S})$ for each of the independently sampled subjects.

To prove the second expression, observe that by the multinomial distribution, we have:

$$\begin{aligned}
&\mathbb{P}\Big(X_c(\mathcal{S},t)\geq 1,\ \forall c\in\mathcal{C}',\ X_g(\mathcal{S},t)=0,\ \forall g\in\mathcal{I}^{\texttt{cml}}(\mathcal{S})\setminus\mathcal{C}'\Big)\\
&\quad=\mathbb{P}\Big(X_c(\mathcal{S},t)\geq 1,\ \forall c\in\mathcal{C}',\ X_g(\mathcal{S},t)=0,\ \forall g\in\mathcal{I}^{\texttt{cml}}(\mathcal{S})\setminus\mathcal{C}',\ X_0(\mathcal{S},t)=t-\sum_{c\in\mathcal{C}'}X_c(\mathcal{S},t)\Big)\\
&\quad=\sum_{\substack{(x_{c_1},x_{c_2},\ldots,x_{c_\ell})\in[t]^\ell:\\ x_{c_1}+x_{c_2}+\ldots+x_{c_\ell}\leq t}}\left[\frac{t!}{\prod_{c\in\mathcal{C}'}x_c!\cdot(t-\sum_{c\in\mathcal{C}'}x_c)!}\cdot\prod_{c\in\mathcal{C}'}(p_c(\mathcal{S}))^{x_c}\cdot(p_0(\mathcal{S}))^{t-\sum_{c\in\mathcal{C}'}x_c}\right].
\end{aligned}$$

Let $x_{\texttt{sum}}(\mathcal{C}):=\sum_{c\in\mathcal{C}}x_c$ for $\mathcal{C}\subseteq\mathcal{C}'$. Re-writing the last equation with the new notation and then switching to the binomial expressions, we get:

$$\begin{aligned}
&=\sum_{\substack{(x_{c_1},x_{c_2},\ldots,x_{c_\ell})\in[t]^\ell:\\ x_{\texttt{sum}}(\mathcal{C}')\leq t}}\left[\frac{t!}{\prod_{c\in\mathcal{C}'}x_c!\cdot(t-x_{\texttt{sum}}(\mathcal{C}'))!}\cdot\prod_{c\in\mathcal{C}'}(p_c(\mathcal{S}))^{x_c}\cdot(p_0(\mathcal{S}))^{t-x_{\texttt{sum}}(\mathcal{C}')}\right]\\
&=\sum_{\substack{(x_{c_1},x_{c_2},\ldots,x_{c_\ell})\in[t]^\ell:\\ x_{\texttt{sum}}(\mathcal{C}')\leq t}}\left[\binom{t}{x_{c_1}}\cdot\binom{t-x_{c_1}}{x_{c_2}}\cdots\binom{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_\ell\})}{x_{c_\ell}}\cdot\prod_{c\in\mathcal{C}'}(p_c(\mathcal{S}))^{x_c}\cdot(p_0(\mathcal{S}))^{t-x_{\texttt{sum}}(\mathcal{C}')}\right],
\end{aligned}$$

which can be expressed as the product of nested summations:

$$\begin{aligned}
&=\sum_{x_{c_1}=1}^{t}\left[\binom{t}{x_{c_1}}\cdot(p_{c_1}(\mathcal{S}))^{x_{c_1}}\cdot\sum_{x_{c_2}=1}^{t-x_{c_1}}\left[\binom{t-x_{c_1}}{x_{c_2}}\cdot(p_{c_2}(\mathcal{S}))^{x_{c_2}}\cdot\ \cdots\right.\right.\\
&\quad\left.\left.\cdot\sum_{x_{c_\ell}=1}^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_\ell\})}\left[\binom{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_\ell\})}{x_{c_\ell}}\cdot(p_{c_\ell}(\mathcal{S}))^{x_{c_\ell}}\cdot(p_0(\mathcal{S}))^{t-x_{\texttt{sum}}(\mathcal{C}')}\right]\right]\right].
\end{aligned}\tag{11}$$

By the binomial theorem, the innermost summation (over x_{c_ℓ}) in (11) can be expressed as:

$$(p_0(\mathcal{S})+p_{c_\ell}(\mathcal{S}))^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_\ell\})}-(p_0(\mathcal{S}))^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_\ell\})}.\tag{12}$$

Substituting (12) into the second innermost summation term (over $x_{c_{\ell-1}}$) in (11), applying the binomial theorem again, and letting $P_{\texttt{sum}}(\mathcal{C}):=p_0(\mathcal{S})+\sum_{c\in\mathcal{C}}p_c(\mathcal{S})$ for $\mathcal{C}\subseteq\mathcal{C}'$ yield:

$$\begin{aligned}
&\sum_{x_{c_{\ell-1}}=1}^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_{\ell-1},c_\ell\})}\binom{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_{\ell-1},c_\ell\})}{x_{c_{\ell-1}}}\cdot(p_{c_{\ell-1}}(\mathcal{S}))^{x_{c_{\ell-1}}}\cdot\left[P_{\texttt{sum}}(\{c_\ell\})^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_\ell\})}-(p_0(\mathcal{S}))^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_\ell\})}\right]\\
&=P_{\texttt{sum}}(\{c_{\ell-1},c_\ell\})^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_{\ell-1},c_\ell\})}-P_{\texttt{sum}}(\{c_\ell\})^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_{\ell-1},c_\ell\})}-P_{\texttt{sum}}(\{c_{\ell-1}\})^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_{\ell-1},c_\ell\})}\\
&+(p_0(\mathcal{S}))^{t-x_{\texttt{sum}}(\mathcal{C}'\setminus\{c_{\ell-1},c_\ell\})}.
\end{aligned}$$

Repeating the same process, e.g., substituting the last derived expression into the previous innermost summation term in (11) and applying the binomial theorem, yields the expression. $\square$

Lemma B.3 (Pool-level retesting set PMF). *Consider any assay $\mathcal{S} \subseteq \mathcal{N}$ with pool size $t \geq 2$. For any disease subset $\mathcal{S}' \subseteq \mathcal{S}$ with $s' = |\mathcal{S}'|$, the probability that the assay's pool-level retesting set, $\mathcal{N}^+_{pool}(\mathcal{S}, t)$, equals set $\mathcal{S}'$ follows:*

$$\mathbb{P}(\mathcal{N}^+_{pool}(\mathcal{S},t) = \mathcal{S}') = (-1)^{s'} \cdot (p_0(\mathcal{S}))^t + \sum_{r=1}^{s'} (-1)^{s'-r} \cdot \left[\sum_{\mathcal{S}'' \in \mathcal{S}'(r)} \left(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}'')} p_c(\mathcal{S}) \right)^t \right].$$

<u>Proof</u> The proof leverages Lemma B.2, which provides various probabilities related to the random vector $\boldsymbol{X}(\mathcal{S},t) = \big(X_0(\mathcal{S},t), (X_c(\mathcal{S},t))_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S})}\big) \sim \text{Multinomial}(t, \boldsymbol{p}(\mathcal{S}))$.

Given $\mathcal{S} \subseteq \mathcal{N}$, consider any $\mathcal{S}' \subseteq \mathcal{S}$ with $s' = |\mathcal{S}'|$. The proof is by induction on s':

Base Cases ($s' = 0$ and $s' = 1$): If $\mathcal{S}' = \emptyset$, then letting $C' = \emptyset$ (with corresponding $\ell = 0$) in Lemma B.2 yields $\mathbb{P}(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \emptyset) = \mathbb{P}(X_c(\mathcal{S},t) = 0,\ \forall c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S})) = \ (p_0(\mathcal{S}))^t$.

Similarly, if $\mathcal{S}' = \{i\}$, $i \in \mathcal{N}$, then letting $C' = i$ (with corresponding $\ell = 1$) in Lemma B.2 yields $\mathbb{P}(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \{i\}) = \mathbb{P}\big(X_i(\mathcal{S},t) \geq 1,\ X_g(\mathcal{S},t) = 0,\ \forall g \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}) \setminus \{i\}\big) = \big(p_0(\mathcal{S}) + p_i(\mathcal{S})\big)^t - (p_0(\mathcal{S}))^t$. For $s = |S| \leq 1$, we are done. For $s \geq 2$, we proceed by considering any $s' \in \{2, \cdots, s\}$ as follows.

Inductive Hypothesis ($< s'$): Assume the lemma holds for all subsets of $\mathcal{S}$ with $|\mathcal{S}| < s'$.

Inductive Step (s'): Let $\mathcal{S}' \subseteq \mathcal{S}$ with $|\mathcal{S}'| = s'$. The event $\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \mathcal{S}'$ indicates: (1) every disease from $\mathcal{S} \setminus \mathcal{S}'$ is absent in the pool, i.e., $X_g(\mathcal{S},t) = 0,\ \forall g \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}) \setminus \mathcal{I}^{\mathtt{cml}}(\mathcal{S}')$ <u>and</u> (2) for every disease $i \in \mathcal{S}'$, at least one category including i appears in the pool, i.e., $\exists c' \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S} \setminus \{i\}) \cup \{0\}$ such that $X_{i \oplus c'}(\mathcal{S},t) \geq 1$. By Lemma B.2, the probability of case (1) is,

$$\mathbb{P}\big(X_g(\mathcal{S},t) = 0,\ \forall g \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}) \setminus \mathcal{I}^{\mathtt{cml}}(\mathcal{S}')\big) = \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}')} p_c(\mathcal{S})\Big)^t,$$

which includes cases where not all diseases in $\mathcal{S}'$ appear in the pool. To ensure case (2) jointly, we apply exclusion over the subsets $\mathcal{S}'' \subset \mathcal{S}'$:

$$\mathbb{P}(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \mathcal{S}') = \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}')} p_c(\mathcal{S})\Big)^t - \mathbb{P}(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \emptyset) - \sum_{r=1}^{s'-1} \sum_{\mathcal{S}'' \in \mathcal{S}'(r)} \mathbb{P}(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \mathcal{S}'').$$

By the inductive hypothesis, for each $\mathcal{S}'' \in \mathcal{S}'(r)$ with $r < s'$, substituting we already know the expressions for $\mathbb{P}(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \mathcal{S}'')$, the substitution of which yields:

$$\begin{aligned} \mathbb{P}(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \mathcal{S}') &= \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}')} p_c(\mathcal{S})\Big)^t - (p_0(\mathcal{S}))^t \\ &\quad - \sum_{r=1}^{s'-1} \sum_{\mathcal{S}'' \in \mathcal{S}'(r)} \Big[(-1)^r (p_0(\mathcal{S}))^t + \sum_{u=1}^{r} (-1)^{r-u} \cdot \sum_{\mathcal{U} \in \mathcal{S}''(u)} \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{U})} p_c(\mathcal{S})\Big)^t \Big]. \end{aligned} \tag{13}$$

We now simplify these terms step-by-step.

$$-\sum_{r=1}^{s'-1} \sum_{\mathcal{S}'' \in \mathcal{S}'(r)} (-1)^r (p_0(\mathcal{S}))^t = -(p_0(\mathcal{S}))^t \cdot \sum_{r=1}^{s'-1} \binom{s'}{r} (-1)^r = [1 + (-1)^{s'}](p_0(\mathcal{S}))^t,$$

the last step using the binomial identity $\sum_{r=0}^{s'} \binom{s'}{r} (-1)^r = 0 \ \Rightarrow \ \sum_{r=1}^{s'-1} \binom{s'}{r} (-1)^r = -1 - (-1)^{s'}$. Thus, combining the $(p_0(\mathcal{S}))^t$-related terms in (13), we obtain $(-1)^{s'} \cdot (p_0(\mathcal{S}))^t$.

For the remaining part in the large summation in (13), we reindex the terms involving $\mathcal{U}$. Originally, we have:

$$-\sum_{r=1}^{s'-1} \sum_{\mathcal{S}'' \in \mathcal{S}'(r)} \sum_{u=1}^{r} (-1)^{r-u} \cdot \sum_{\mathcal{U} \in \mathcal{S}''(u)} \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{U})} p_c(\mathcal{S})\Big)^t, \tag{14}$$

where $\mathcal{S}''$ comes from r-subsets of $\mathcal{S}'$, and $\mathcal{U}$ comes from u-subsets of $\mathcal{S}''$. To simplify this, we *reindex* by focusing directly on the u-subsets of $\mathcal{S}'$ where $u \in \{1,2,\dots,s'-1\}$. After reindexing, we first fix u and consider all $\mathcal{U} \in \mathcal{S}'(u)$. We then account for how $\mathcal{U}$ arises as a subset of $\mathcal{S}''$ which has various sizes $r \geq u$. After performing this reindexing, (14) can be simplified to:

$$\sum_{u=1}^{s'-1} (-1)^{s'-u} \cdot \sum_{\mathcal{U} \in \mathcal{S}'(u)} \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{U})} p_c(\mathcal{S})\Big)^t. \tag{15}$$

We next discuss how the signs $(-1)^{s'-u}$ in (15) are obtained. Consider (14) and fix a u-subset $\mathcal{U} \subseteq \mathcal{S}'$. Its contribution to the summation comes from all subsets $\mathcal{S}'' \subseteq \mathcal{S}'$ of size r with $\mathcal{U} \subseteq \mathcal{S}''$ and $u \leq r \leq s'-1$. Each such subset $\mathcal{S}''$ adds a sign $(-1)^{r-u}$, thus the total contribution for $\mathcal{U}$ is:

$$\sum_{r=u}^{s'-1} (-1)^{r-u} \cdot \binom{s'-u}{r-u} \overset{\text{change of variable}}{=} \sum_{m=0}^{s'-u-1} (-1)^m \cdot \binom{s'-u}{m} \overset{\text{binomial theorem}}{=} -(-1)^{s'-u}.$$

Putting all the parts together (replacing u and $\mathcal{U}$ in (15) with r and $\mathcal{S}''$, resp.), we obtain:

$$\begin{aligned}
(13) &= (-1)^{s'} \cdot (p_0(\mathcal{S}))^t + \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}')} p_c(\mathcal{S})\Big)^t + \sum_{r=1}^{s'-1} (-1)^{s'-r} \cdot \sum_{\mathcal{S}'' \in \mathcal{S}'(r)} \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}'')} p_c(\mathcal{S})\Big)^t \\
&= (-1)^{s'} \cdot (p_0(\mathcal{S}))^t + \sum_{r=1}^{s'} (-1)^{s'-r} \cdot \sum_{\mathcal{S}'' \in \mathcal{S}'(r)} \Big(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\mathtt{cml}}(\mathcal{S}'')} p_c(\mathcal{S})\Big)^t, \text{ the desired formula.} \quad \square
\end{aligned}$$

Lemma B.4 (Pool-level retesting set cardinality). *For any given assay $\mathcal{S} \subseteq \mathcal{N}$ with $s = |\mathcal{S}|$ and pool size $t \geq 2$, $\mathbb{E}[D_{pool}(\mathcal{S},t)] = s - \sum_{i \in \mathcal{S}} (1-\pi_i)^t$.*

Proof The proof follows by construction, using the PMF of $\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t)$ from Lemma B.3. We consider cases based on the size the assay (s). Note that if $\mathcal{S} = \emptyset$, the result trivially holds.

- Consider any singleton assay $\mathcal{S} = \{i\}, i \in \mathcal{N}$. We have:

$$\begin{aligned}
\mathbb{E}[D_{\mathtt{pool}}(\mathcal{S}=\{i\},t)] &\overset{\text{definition}}{=} 1 \cdot \underbrace{\mathbb{P}\left(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \{i\}\right)} + 0 \cdot \left(1 - \mathbb{P}\left(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \{i\}\right)\right) \\
&\overset{\text{Lemma B.3}}{=} (\underbrace{p_0(\mathcal{S}) + p_i(\mathcal{S})}_{=1})^t - (\underbrace{p_0(\mathcal{S})}_{=1-\pi_i})^t \overset{\text{Lemma B.1}}{=} 1 - (1-\pi_i)^t.
\end{aligned}$$

- Consider any assay $\mathcal{S} = \{i,j\}$ where $i \in \mathcal{N}, j \in \mathcal{N} \setminus \{i\}$. We have:

$$\begin{aligned}
\mathbb{E}[D_{\mathtt{pool}}(\mathcal{S}=\{i,j\},t)] &\overset{\text{definition}}{=} 1 \cdot \left[\mathbb{P}\left(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \{i\}\right) + \mathbb{P}\left(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \{j\}\right)\right] \\
&\quad + 2 \cdot \mathbb{P}\left(\mathcal{N}^+_{\mathtt{pool}}(\mathcal{S},t) = \{i,j\}\right) \\
&\overset{\text{Lemma B.3}}{=} (p_0(\mathcal{S}) + p_i(\mathcal{S}))^t - \cancel{(p_0(\mathcal{S}))^t} + (p_0(\mathcal{S}) + p_j(\mathcal{S}))^t - \cancel{(p_0(\mathcal{S}))^t} \\
&\quad + 2 \cdot \Big(\underbrace{p_0(\mathcal{S}) + p_i(\mathcal{S}) + p_j(\mathcal{S}) + p_{i \oplus j}(\mathcal{S})}_{=1 \text{ by definition since } \mathcal{S}=\{i,j\}}\Big)^t - 2 \cdot \Big(\underbrace{p_0(\mathcal{S}) + p_i(\mathcal{S})}_{1-\pi_j}\Big)^t - 2 \cdot \Big(\underbrace{p_0(\mathcal{S}) + p_j(\mathcal{S})}_{1-\pi_i}\Big)^t \\
&\quad \cancel{+ 2 \cdot (p_0(\mathcal{S}))^t} \overset{\text{Lemma B.1}}{=} 2 - (1-\pi_j)^t - (1-\pi_i)^t,
\end{aligned}$$

where in the last step we use Lemma B.1, which yields, for instance, $\pi_i = p_i(\mathcal{S}) + p_{i \oplus j}(\mathcal{S})$ and combine it with $p_0(\mathcal{S}) + p_j(\mathcal{S}) = 1 - (p_i(\mathcal{S}) + p_{i \oplus j}(\mathcal{S}))$ for this special case of $\mathcal{S} = \{i,j\}$.

- Now, consider the general case with $s \geq 3$. We have:

$$
\begin{aligned}
\mathbb{E}\left[D_{\texttt{pool}}(\mathcal{S},t)\right] &\overset{\text{definition}}{=} \sum_{r=1}^{s} \sum_{\mathcal{S}' \in \mathcal{S}(r)} r \cdot \mathbb{P}\left(\mathcal{N}^{+}_{\texttt{pool}}(\mathcal{S},t) = \mathcal{S}'\right) \\
&\overset{\text{Lemma B.3}}{=} \sum_{r=1}^{s} r \cdot \sum_{\mathcal{S}' \in \mathcal{S}(r)} \left(\cancel{(-1)^{s'} \cdot (p_0(\mathcal{S}))^t} + \sum_{r'=1}^{s'} (-1)^{s'-r'} \cdot \left[\sum_{\mathcal{S}'' \in \mathcal{S}'(r')} \left(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S}'')} p_c(\mathcal{S}) \right)^t \right] \right) \\
&\overset{\text{re-arrange}}{=} (p_0(\mathcal{S}))^t \cdot \underbrace{\left(\sum_{r=1}^{s} r \cdot \sum_{\mathcal{S}' \in \mathcal{S}(r)} (-1)^{s'} \right)}_{=0 \text{ by (i)}} \\
&\quad + \sum_{r=1}^{s-2} \sum_{\hat{\mathcal{S}} \in \mathcal{S}(r)} \left(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\texttt{cml}}(\hat{\mathcal{S}})} p_c(\mathcal{S}) \right)^t \cdot \underbrace{\left(\sum_{r'=\hat{s}}^{s} r' \cdot \sum_{\mathcal{S}' \in \mathcal{S}(r') \,:\, \hat{\mathcal{S}} \subseteq \mathcal{S}'} (-1)^{r'-\hat{s}} \right)}_{=0 \text{ by (ii)}} \\
&\quad + \sum_{\hat{\mathcal{S}} \in \mathcal{S}(s-1)} \underbrace{\left(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\texttt{cml}}(\hat{\mathcal{S}})} p_c(\mathcal{S}) \right)^t}_{\substack{=1-\pi_{\mathcal{I}(\mathcal{S}\setminus\hat{\mathcal{S}})} \text{ by Lemma B.1} \\ \text{combined with } p_0(\mathcal{S})+\pi(\mathcal{S})=1}} \cdot \underbrace{\left(\sum_{r'=s-1}^{s} r' \cdot \sum_{(\text{unique})\mathcal{S}' \in \mathcal{S}(r') \,:\, \hat{\mathcal{S}} \subseteq \mathcal{S}'} (-1)^{r'-(s-1)} \right)}_{=(s-1)\cdot(-1)^0 + s\cdot(-1)^1 = s-1-s = -1} \\
&\quad + s \cdot \underbrace{\left(p_0(\mathcal{S}) + \sum_{c \in \mathcal{I}^{\texttt{cml}}(\mathcal{S})} p_c(\mathcal{S}) \right)^t}_{=1 \text{ by definition}},
\end{aligned}
$$

which yields the result of $s - \sum_{i \in \mathcal{S}} (1-\pi_i)^t$. We now provide the detailed arguments for (i)-(ii).
Derivation of (i): First, observe that the expression is equal to $\sum_{r=1}^{s} r \cdot \binom{s}{r} \cdot (-1)^r$. Using the combinatorial identity $r \cdot \binom{s}{r} = s \cdot \binom{s-1}{r-1}$, the summation becomes: $s \cdot \sum_{r=1}^{s} \binom{s-1}{r-1} \cdot (-1)^r$. By substituting $k = r-1$, so that $r = k+1$, we rewrite it as:

$$
s \cdot \sum_{r=1}^{s} \binom{s-1}{r-1} \cdot (-1)^r = s \cdot \sum_{k=0}^{s-1} \binom{s-1}{k} \cdot (-1)^{k+1} = -s \cdot \sum_{k=0}^{s-1} \binom{s-1}{k} \cdot (-1)^k \overset{\substack{\text{binomial} \\ \text{theorem}}}{=} 0.
$$

Derivation of (ii): We proceed by analyzing the inner summation. For a fixed r', the subsets $\mathcal{S}' \in \mathcal{S}(r')$ that contain $\hat{\mathcal{S}}$ can be constructed by fixing all elements of $\hat{\mathcal{S}}$ as elements of $\mathcal{S}'$ and selecting the remaining $r' - \hat{s}$ elements from $\mathcal{S} \setminus \hat{\mathcal{S}}$. Thus, there are $\binom{s-\hat{s}}{r'-\hat{s}}$ such $\mathcal{S}'$ subsets, and each subset contributes $(-1)^{r'-\hat{s}}$ to the inner summation. Therefore, we get:

$$
\sum_{\mathcal{S}' \in \mathcal{S}(r') : \hat{\mathcal{S}} \subseteq \mathcal{S}'} (-1)^{r'-\hat{s}} = \binom{s-\hat{s}}{r'-\hat{s}} \cdot (-1)^{r'-\hat{s}}.
$$

Substituting this into the original summation, we obtain:

$$
\sum_{r'=\hat{s}}^{s} r' \cdot \sum_{\mathcal{S}' \in \mathcal{S}(r') : \hat{\mathcal{S}} \subseteq \mathcal{S}'} (-1)^{r'-\hat{s}} = \sum_{r'=\hat{s}}^{s} r' \cdot \binom{s-\hat{s}}{r'-\hat{s}} \cdot (-1)^{r'-\hat{s}}.
$$

Change of index as $k = r' - \hat{s}$ (i.e., $r' = k + \hat{s}$) yields:

$$
\sum_{k=0}^{s-\hat{s}} (k+\hat{s}) \binom{s-\hat{s}}{k} (-1)^k = \sum_{k=0}^{s-\hat{s}} k \binom{s-\hat{s}}{k} (-1)^k + \hat{s} \sum_{k=0}^{s-\hat{s}} \binom{s-\hat{s}}{k} (-1)^k.
$$

By the binomial theorem, we have $\sum_{k=0}^{s-\hat{s}}\binom{s-\hat{s}}{k}(-1)^k=(1-1)^{s-\hat{s}}=0$, thus the second term is 0. To show that $\sum_{k=0}^{s-\hat{s}}k\binom{s-\hat{s}}{k}(-1)^k=0$, consider $(1+x)^{s-\hat{s}}=\sum_{k=0}^{s-\hat{s}}\binom{s-\hat{s}}{k}x^k$. Differentiating both sides with respect to x, we get $(s-\hat{s})(1+x)^{s-\hat{s}-1}=\sum_{k=0}^{s-\hat{s}}k\binom{s-\hat{s}}{k}x^{k-1}$. Multiplying both sides by x yields $x(s-\hat{s})(1+x)^{s-\hat{s}-1}=\sum_{k=0}^{s-\hat{s}}k\binom{s-\hat{s}}{k}x^k$. Setting $x=-1$, we have $(-1)(s-\hat{s})(1-1)^{s-\hat{s}-1}=\sum_{k=0}^{s-\hat{s}}k\binom{s-\hat{s}}{k}(-1)^k$. The LHS is 0, thus the RHS must also be 0. Since both parts vanish, the entire summation is 0, which concludes the proof. □

B.3.2. Main Results

Proof of Theorem 1 *Part 1.* $D_{\texttt{subject}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})=0$ iff the random subject does not have any diseases from set $\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$, which happens with probability $p_0(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}))$, and all pool outcomes the random subject is included in are negative considering the remaining subjects:

$$\begin{aligned}\mathbb{P}\left(D_{\texttt{subject}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})=0\right)&=p_0(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}))\cdot\mathbb{P}\Big(\bigcap_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\{\mathcal{N}^{+}_{\texttt{pool}}(\mathcal{S}_k,t_k-1)=\emptyset\}\Big)\\&=p_0(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}))\cdot\prod_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\left(p_0(\mathcal{S}_k)\right)^{t_k-1},\end{aligned}$$

where the last equality follows by Lemma B.3, since assay pools are constructed independently.
For the first moment of $D_{\texttt{subject}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$, $\mathcal{N}(c)$ is the set of diseases in category c, with cardinality $n_c:=|\mathcal{N}(c)|$. Conditioning on the random subject's disease category and using the law of total expectation and the first moment of $D_{\texttt{pool}}(\mathcal{S}_k\setminus\mathcal{N}(c),t_k-1)$ (Lemma B.4):

$$\mathbb{E}\left[D_{\texttt{subject}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right]=\sum_{c\in\{0\}\cup\mathcal{I}^{\texttt{cml}}\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)}p_c\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\cdot\left[n_c+\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\mathbb{E}\left[D_{\texttt{pool}}(\mathcal{S}_k\setminus\mathcal{N}(c),t_k-1)\right]\right],$$

that is, with probability $p_c\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)$, the (fixed) random subject will need retesting for the n_c diseases they are positive for, independently of the disease status of the subjects they are pooled with. Further, some other diseases might be added to this subject's retesting set based on the disease status of the subjects they are pooled with in the various assay pools, which we express by (i) removing the diseases this subject is positive for from their respective pooled assays, and (ii) reducing their pool sizes by one (due to the removal of the conditioned random subject). Then, substituting the expressions from Lemma B.4,

$$\begin{aligned}\mathbb{E}\left[D_{\texttt{subject}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right]&=\sum_{c\in\{0\}\cup\mathcal{I}^{\texttt{cml}}\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)}p_c\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\cdot\left[n_c+\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\left(|\mathcal{S}_k\setminus\mathcal{N}(c)|-\sum_{i\in\mathcal{S}_k\setminus\mathcal{N}(c)}(1-\pi_i)^{t_k-1}\right)\right]\\&=\underbrace{\sum_{c\in\{0\}\cup\mathcal{I}^{\texttt{cml}}(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}))}p_c\Big(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\Big)}_{=1\text{ by definition}}\cdot\Big[\underbrace{n_c+\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\left(|\mathcal{S}_k\setminus\mathcal{N}(c)|\right.}_{n_c+(n_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})-n_c)}\Big]\\&\quad-\sum_{c\in\{0\}\cup\mathcal{I}^{\texttt{cml}}(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}))}p_c\Big(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\Big)\cdot\sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\setminus\mathcal{N}_c}(1-\pi_i)^{w_i(\boldsymbol{t})-1}\\&=n_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})-\sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(1-\pi_i)^{w_i(\boldsymbol{t})-1}\cdot\sum_{c\in\{0\}\cup\mathcal{I}^{\texttt{cml}}\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\setminus\{i\}\right)}p_c\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\\&=n_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})-\sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(1-\pi_i)^{w_i(\boldsymbol{t})-1}\cdot\left[1-\underbrace{\left(1-\sum_{c\in\{0\}\cup\mathcal{I}^{\texttt{cml}}\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\setminus\{i\}\right)}p_c\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\right)}_{(\star)}\right].\end{aligned}$$

$$\text{Note that } (\star) = \sum_{c\in\{0\}\cup\mathcal{I}^{\texttt{cml}}\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)} p_c\big(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\big) - \sum_{c\in\{0\}\cup\mathcal{I}^{\texttt{cml}}\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\setminus\{i\}\right)} p_c\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)$$
$$= p_i(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})) + \sum_{c\in\mathcal{I}^{\texttt{cml}}(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\setminus\{i\})} p_{i\oplus c}(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})) = \pi_i,$$

where the last step is obtained from Lemma B.1 using $\mathcal{S}=\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$. Substituting π_i into $(\star)$ in the last equation yields the desired expression.

Part 2. By part 1 of this result, the adaptive cost expression in (3) reduces to:

$$\begin{aligned}
C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}) = &\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})} (c_f+s_k\cdot c_v)\cdot\frac{1}{t_k} + c_f\cdot\Big(1-p_0\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\cdot\prod_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\big(p_0(\mathcal{S}_k)\big)^{t_k-1}\Big)\\
&+c_v\cdot\Big(n_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}) - \sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(1-\pi_i)^{w_i(\boldsymbol{t})}\Big) + \sum_{k\in\mathcal{K}_{\texttt{ind}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(c_f+s_k\cdot c_v)\\
=\ & c_f\cdot\Big(\big(\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\frac{1}{t_k}\big)+1-p_0\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\cdot\prod_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\big(p_0(\mathcal{S}_k)\big)^{t_k-1}\Big)\\
&+c_v\cdot\Big(\sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\Big[\frac{1}{w_i(\boldsymbol{t})}+1-(1-\pi_i)^{w_i(\boldsymbol{t})}\Big]\Big) + \sum_{k\in\mathcal{K}_{\texttt{ind}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(c_f+s_k\cdot c_v),
\end{aligned}$$

where in the transition we used $\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(s_k/t_k) = \sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(1/w_i(\boldsymbol{t}))$ and $n_{\texttt{pool}} = \sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})} 1.$

Now, we show that $C^{\texttt{Dorf}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}) - C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}) \geq 0$ for any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$ and $\boldsymbol{t}$. By (4) and (5), the cost difference is:

$$\begin{aligned}
&c_f\cdot\Big(\Big[\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}[1-(1-\pi(\mathcal{S}_k))^{t_k}]\Big]-1+p_0\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\cdot\prod_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\big(p_0(\mathcal{S}_k)\big)^{t_k-1}\Big)+\\
&c_v\cdot\Big(\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\big[\frac{s_k}{t_k}+s_k\big] - \sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\big[\frac{1}{w_i(\boldsymbol{t})}+1\big] - \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\big[s_k\cdot(1-\pi(\mathcal{S}_k))^{t_k}\big] + \sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(1-\pi_i)^{w_i(\boldsymbol{t})}\Big).
\end{aligned}\tag{16}$$

It is sufficient to show that the terms in parenthesis multiplying c_f and c_v are nonnegative. Noting that $\pi(\mathcal{S})\geq\pi_i, i\in\mathcal{S}$, the latter directly follows from the following two relationships:

$$\begin{aligned}
&\text{(i)}\quad \sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\big[\frac{1}{w_i(\boldsymbol{t})}+1\big] = \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\sum_{i\in\mathcal{S}_k}\big[\frac{1}{w_i(\boldsymbol{t})}+1\big] = \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\big[\frac{s_k}{t_k}+s_k\big]\\
&\text{(ii)}\quad \sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(1-\pi_i)^{w_i(\boldsymbol{t})} = \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\sum_{i\in\mathcal{S}_k}(1-\pi_i)^{t_k}\\
&\qquad\qquad \geq \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\sum_{i\in\mathcal{S}_k}(1-\pi(\mathcal{S}_k))^{t_k} = \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})} s_k\cdot(1-\pi(\mathcal{S}_k))^{t_k}.
\end{aligned}$$

To analyze the term multiplying c_f, we first define:

$$\Psi(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}) := \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}(1-\pi(\mathcal{S}_k))^{t_k} - p_0\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\cdot\prod_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\big(p_0(\mathcal{S}_k)\big)^{t_k-1},\ \text{where}$$
$$\frac{\partial\Psi(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}{\partial t_k} = (\underbrace{1-\pi(\mathcal{S}_k)}_{=p_0(\mathcal{S}_k)})^{t_k}\cdot\ln(\underbrace{1-\pi(\mathcal{S}_k)}_{=p_0(\mathcal{S}_k)})$$

$$
\begin{aligned}
&- (p_0(\mathcal{S}_k))^{t_k-1}\cdot \ln(p_0(\mathcal{S}_k))\cdot p_0\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\cdot \prod_{k'\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\setminus\{k\}} (p_0(\mathcal{S}_{k'}))^{t_{k'}-1}\\
&= \underbrace{\ln(p_0(\mathcal{S}_k))}_{\le 0}\cdot\underbrace{(p_0(\mathcal{S}_k))^{t_k-1}}_{\ge 0}\cdot\Bigg[\overbrace{p_0(\mathcal{S}_k) - \underbrace{p_0\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\cdot \prod_{k'\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\setminus\{k\}} (p_0(\mathcal{S}_{k'}))^{t_{k'}-1}}_{\le\ p_0\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right)\ \le\ p_0(\mathcal{S}_k)}}^{\ge 0}\Bigg].
\end{aligned}
$$

Thus, $\Psi(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$ is nonincreasing in t_k. Therefore, we have:

$$
\Psi(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}) \le \Psi(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\mathbf{1}) = |\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})| - \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\pi(\mathcal{S}_k) - p_0\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right),
$$

using which we obtain that the term multiplying c_f in (16) is equivalent to:

$$
\begin{aligned}
&|\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})| - 1 - \Psi(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}) \ge\ |\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})| - 1 - \Psi(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\mathbf{1})\\
&= \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\pi(\mathcal{S}_k) + \left(p_0\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right) - 1\right) = \sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\pi(\mathcal{S}_k) - \pi\left(\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})\right) \ge 0.
\end{aligned}
$$

The last inequality follows because the collection of sets $\mathcal{S}_k, k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$, forms a partition of $\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$, i.e., $\cup_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})}\mathcal{S}_k = \mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$. $\square$

B.4. Proofs for §3.2

$I_q := qc_f + nc_v$ denotes the all-assay individual testing cost for any q-partition portfolio, and

$$
C(\mathcal{S},t;\ \mathcal{S}\text{-only}) := \frac{c_f + sc_v}{t} + c_f\left(1-\gamma(\mathcal{S})^t\right) + c_v\left(s - \sum_{i\in\mathcal{S}}(1-\pi_i)^t\right),\quad \forall \mathcal{S}\subseteq\mathcal{N}, \tag{17}
$$

that is, the adaptive cost for assay $\mathcal{S}$ evaluated *in isolation*, treating $\mathcal{S}$ as if it constituted the complete disease set. Thus, $C(\mathcal{N},t;\ \mathcal{N}\text{-only})$ recovers the n-plex cost $C\left(\boldsymbol{\mathcal{S}}^{n\text{-plex}},t\right)$.

B.4.1. Supporting Result

Lemma B.5 (Link between adaptive threshold and cost). *For any assay $\mathcal{S}\subseteq\mathcal{N}$, $\pi(\mathcal{S}) < \underline{p}(\mathcal{S})$ iff there exists $t\ge 2$ such that $C(\mathcal{S},t;\ \mathcal{S}\text{-only}) < c_f + sc_v$.*

<u>Proof</u> Because $\pi(\mathcal{S})\in(0,1)$, we have that $\gamma(\mathcal{S}) = 1-\pi(\mathcal{S})\in(0,1)$.
($\Leftarrow$) Given that $\exists t\ge 2 : C(\mathcal{S},t;\ \mathcal{S}\text{-only}) < c_f + sc_v$, we apply (17) and rearrange terms:

$$
\begin{aligned}
&\tfrac{c_f+sc_v}{t} + c_f(1-\gamma(\mathcal{S})^t) + c_v\left(s - \textstyle\sum_{i\in\mathcal{S}}(1-\pi_i)^t\right) < c_f + sc_v\\
\iff\ & 1-\gamma(\mathcal{S})^t < \left(1+\tfrac{sc_v}{c_f}\right) - \tfrac{1}{t}\left(1+\tfrac{sc_v}{c_f}\right) - \tfrac{sc_v}{c_f} + \tfrac{c_v}{c_f}\textstyle\sum_{i\in\mathcal{S}}(1-\pi_i)^t\\
\iff\ & 1-\gamma(\mathcal{S})^t < 1 - \left[\tfrac{1}{t}\left(1+\tfrac{sc_v}{c_f}\right) - \tfrac{c_v}{c_f}\textstyle\sum_{i\in\mathcal{S}}(1-\pi_i)^t\right]\\
\iff\ & \gamma(\mathcal{S})^t > \rho(\mathcal{S},t).
\end{aligned} \tag{18}
$$

If $\rho(\mathcal{S},t)\ge 1$, then (18) cannot hold for this t, as $\gamma(\mathcal{S}) < 1$. Therefore, we must have $\rho(\mathcal{S},t) < 1$. Then, noting that $\gamma(\mathcal{S}) = 1-\pi(\mathcal{S}) > 0$, (18) implies:

$$
\gamma(\mathcal{S}) > (\rho(\mathcal{S},t))^+)^{1/t} \iff \pi(\mathcal{S}) < 1 - ((\rho(\mathcal{S},t))^+)^{1/t} = p^*(\mathcal{S},t)\quad (\text{since } \rho(\mathcal{S},t) < 1),
$$

where $\pi(\mathcal{S}) < p^*(\mathcal{S},t) \le \max_{t'\ge 2} p^*(\mathcal{S},t') = \underline{p}(\mathcal{S})$ (by Definition 2), and the result holds.
($\Rightarrow$) Conversely, if $\pi(\mathcal{S}) < \underline{p}(\mathcal{S}) = \max_{t'\ge 2} p^*(\mathcal{S},t')$, then by definition $\exists t \ge 2$ such that $\pi(\mathcal{S}) < p^*(\mathcal{S},t)$. Since $\pi(\mathcal{S}) > 0$, this implies $p^*(\mathcal{S},t) > 0$, and thus $\rho(\mathcal{S},t) < 1$. The inequality $\pi(\mathcal{S}) < 1 - ((\rho(\mathcal{S},t))^+)^{1/t}$ rearranges to $\gamma(\mathcal{S})^t > (\rho(\mathcal{S},t))^+ \ge \rho(\mathcal{S},t)$. Reversing the algebraic steps in (18) confirms that $C(\mathcal{S},t;\ \mathcal{S}\text{-only}) < c_f + sc_v$ for this t. □

B.4.2. Main Results

Proof of Theorem 2 *Part 1.* By Lemma B.5, $\pi(\mathcal{N}) < \underline{p}(\mathcal{N})$ (the given condition) iff $\exists t \ge 2$ such that $C(\mathcal{N},t;\ \mathcal{N}\text{-only}) < I_1 = c_f + nc_v$. Then the result follows because $C(\mathcal{N},t;\ \mathcal{N}\text{-only}) = C(\boldsymbol{\mathcal{S}}^{n\text{-}\mathtt{plex}},t)$.
Part 2. The proof follows by contradiction. Consider any $\boldsymbol{\mathcal{S}}^{\mathtt{Portfolio}}$. Assume there exists an optimal pool size vector $\boldsymbol{t}^*$ where some assay $\mathcal{S}_k$ is tested individually ($t_k^* = 1$) while others follow $\boldsymbol{t}^*_{-k}$. Wlog, assume all other assays are pooled ($\boldsymbol{t}^*_{-k} \ge \mathbf{2}$), because any other assay tested individually contributes an independent term to the cost and can be excluded from the analysis without affecting the optimal pool sizes for the remaining assays. We compare this solution to an alternative solution $\boldsymbol{t}'$ where $\mathcal{S}_k$ is pooled with size $t_k' \ge 2$, and all other pool sizes remain unchanged ($\boldsymbol{t}'_{-k} = \boldsymbol{t}^*_{-k}$):

$$C(\boldsymbol{\mathcal{S}}^{\mathtt{Portfolio}},\boldsymbol{t}^*) = (c_f + s_k c_v) + \sum_{\ell\ne k} \frac{c_f + s_\ell c_v}{t_\ell^*} + c_v \sum_{\ell \ne k}\left(s_\ell - \sum_{i\in\mathcal{S}_\ell}(1-\pi_i)^{t_\ell^*}\right)$$
$$+ c_f\left(1 - p_0(\mathcal{N}\setminus\mathcal{S}_k)\cdot \prod_{\ell\ne k}(1-\pi(\mathcal{S}_\ell))^{t_\ell^*-1}\right),$$

$$C(\boldsymbol{\mathcal{S}}^{\mathtt{Portfolio}},\boldsymbol{t}') = \frac{c_f + s_k c_v}{t_k'} + \sum_{\ell\ne k}\frac{c_f + s_\ell c_v}{t_\ell^*} + c_v\left(\left(s_k - \sum_{i\in\mathcal{S}_k}(1-\pi_i)^{t_k'}\right) + \sum_{\ell\ne k}\left(s_\ell - \sum_{i\in\mathcal{S}_\ell}(1-\pi_i)^{t_\ell^*}\right)\right)$$
$$+ c_f\left(1 - p_0\cdot(1-\pi(\mathcal{S}_k))^{t_k'-1}\cdot\prod_{\ell\ne k}(1-\pi(\mathcal{S}_\ell))^{t_\ell^*-1}\right).$$

Let $\Delta_k := C(\boldsymbol{\mathcal{S}}^{\mathtt{Portfolio}},\boldsymbol{t}') - C(\boldsymbol{\mathcal{S}}^{\mathtt{Portfolio}},\boldsymbol{t}^*)$. By canceling terms common to assays $\ell \ne k$:

$$\Delta_k = \left[\frac{c_f + s_k c_v}{t_k'} + c_v\left(s_k - \sum_{i\in\mathcal{S}_k}(1-\pi_i)^{t_k'}\right) - (c_f + s_k c_v)\right] + c_f\left(P_{neg}(\boldsymbol{t}^*) - P_{neg}(\boldsymbol{t}')\right), \tag{19}$$

where $Q = \prod_{\ell\ne k}(1-\pi(\mathcal{S}_\ell))^{t_\ell^*-1}$, $\quad P_{neg}(\boldsymbol{t}^*) = p_0(\mathcal{N}\setminus\mathcal{S}_k)\cdot Q$, $\quad P_{neg}(\boldsymbol{t}') = p_0\cdot\gamma(\mathcal{S}_k)^{t_k'-1}\cdot Q$.
As $\pi(\mathcal{S}_k) < \underline{p}(\mathcal{S}_k)$ (the given condition), by Lemma B.5, $\exists t_k' \ge 2 : C(\mathcal{S}_k, t_k';\ \mathcal{S}_k\text{-only}) < c_f + s_k c_v$, equivalently, by (17):

$$\frac{c_f + s_k c_v}{t_k'} \;+\; c_f\left(1 - \gamma(\mathcal{S}_k)^{t_k'}\right) \;+\; c_v\left(s_k - \sum_{i\in\mathcal{S}_k}(1-\pi_i)^{t_k'}\right) < c_f + s_k c_v.$$

Substituting this inequality into (19):

$$\Delta_k < -c_f(1-\gamma(\mathcal{S}_k)^{t_k'}) + c_f(P_{neg}(\boldsymbol{t}^*) - P_{neg}(\boldsymbol{t}')). \tag{20}$$

In the following we show that $\Delta_k < 0$. First, note the following probability bounds:

$$p_0(\mathcal{N}\setminus\mathcal{S}_k) \le p_0 + \pi(\mathcal{S}_k), \quad p_0 \le \gamma(\mathcal{S}_k) \text{ (since } \pi(\mathcal{S}_k)\le\pi(\mathcal{N})).$$

Examining the probability difference term,

$$\begin{aligned} P_{neg}(\boldsymbol{t}^*) - P_{neg}(\boldsymbol{t}') &= Q\cdot\left[p_0(\mathcal{N}\setminus\mathcal{S}_k) - p_0\cdot\gamma(\mathcal{S}_k)^{t_k'-1}\right] \\ &\le 1\cdot\left[p_0 + \pi(\mathcal{S}_k) - p_0\cdot\gamma(\mathcal{S}_k)^{t_k'-1}\right] \qquad (\text{since } Q\le 1) \\ &= \pi(\mathcal{S}_k) + p_0(1-\gamma(\mathcal{S}_k)^{t_k'-1}) \end{aligned}$$

$$
\begin{aligned}
&\le 1-\gamma(\mathcal{S}_k)+\gamma(\mathcal{S}_k)(1-\gamma(\mathcal{S}_k)^{t'_k-1})\\
&=1-\gamma(\mathcal{S}_k)+\gamma(\mathcal{S}_k)-\gamma(\mathcal{S}_k)^{t'_k}=1-\gamma(\mathcal{S}_k)^{t'_k},
\end{aligned}
$$

which implies, by (20), that $\Delta_k<0$. This contradicts the assumption that $t_k^*=1$ is optimal, proving that in an optimal solution assay $\mathcal{S}_k$ is pooled. Further, the result $\Delta_k<0$ holds for all possible prevalences of the other assays $\pi(\mathcal{S}_\ell),\ell\neq k$, which only influence Δ_k through the term Q, and $Q\le 1$ always maintains $\Delta_k<0$. Therefore, the optimality of pooling for $\mathcal{S}_k$ holds for any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$.

Part 3. We wish to show that if $\pi(\mathcal{S})<\underline{p}^{\texttt{Dorf}}$, then $\pi(\mathcal{S})<\underline{p}(\mathcal{S}),\forall\mathcal{S}\subseteq\mathcal{N}$.

We have, $\pi(\mathcal{S})<\underline{p}^{\texttt{Dorf}} \iff \exists t\ge 2:(1-\pi(\mathcal{S}))^t>1/t$ (from (4) in light of Remark 1, Proposition B.1, and Property B.1). Then, noting that $\gamma(\mathcal{S})=1-\pi(\mathcal{S})$, by (18), it suffices to show that:

$$
(1-\pi(\mathcal{S}))^t+\frac{c_v}{c_f}\sum_{i\in\mathcal{S}}(1-\pi_i)^t>\frac{1}{t}\left(1+\frac{sc_v}{c_f}\right).
$$

Since $1-\pi_i\ge 1-\pi(\mathcal{S})$, we have $\sum_{i\in\mathcal{S}}(1-\pi_i)^t\ge s(1-\pi(\mathcal{S}))^t$. Then, because $1-\pi(\mathcal{S}))^t>1/t$,

$$
(1-\pi(\mathcal{S}))^t+\frac{c_v}{c_f}\sum_{i\in\mathcal{S}}(1-\pi_i)^t\ \ge\ (1-\pi(\mathcal{S}))^t\left(1+\frac{sc_v}{c_f}\right)>\frac{1}{t}\left(1+\frac{sc_v}{c_f}\right),\ \forall\mathcal{S}\subseteq\mathcal{N}.
$$

Therefore, $\underline{p}^{\texttt{Dorf}}\le\underline{p}(\mathcal{S}),\ \forall\mathcal{S}\subseteq\mathcal{N}$. ☐

Proof of Proposition 1 *Part 1.* We have, $I_q=I_1+(q-1)c_f,\ \forall q\ge 2$. Assume $\exists t\ge 2$ such that $C(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}},t)<I_1=c_f+nc_v$ (the given condition). In the following, for any q-partition $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$, $q\in[n]$, we construct a pool size solution such that at least one assay is pooled and its cost is lower than the all-assay individual testing solution (i.e., $C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})<I_q$), concluding that an all-individual testing solution ($\boldsymbol{t}=\boldsymbol{1}$) cannot be optimal for $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$.

We do this by constructing a pool size solution in which all assays in the portfolio use some *common* pool size $t\ge 2$. Let $C_{var}(t)$ denote the stage 2 variable cost for this solution:

$$
C_{var}(t)=c_v\left(n-\sum_{i=1}^{n}(1-\pi_i)^t\right),
$$

and the cost functions follow:

$$
\begin{aligned}
C(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}},t)&=\frac{c_f+nc_v}{t}+c_f\left[1-P_{neg}(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}},t)\right]+C_{var}(t),\\
C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},t)&=\sum_{k\in[q]}\frac{c_f+s_kc_v}{t}+c_f\left[1-P_{neg}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},t)\right]+C_{var}(t),\\
\text{where } P_{neg}(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}},t)&=p_0^t,\quad P_{neg}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},t)=p_0\cdot\prod_{k\in[q]}(1-\pi(\mathcal{S}_k))^{t-1}.
\end{aligned}\tag{21}
$$

Noting that $\sum_{k\in[q]}\frac{c_f+s_kc_v}{t}=\frac{qc_f+nc_v}{t}=\frac{c_f+nc_v}{t}+\frac{(q-1)c_f}{t}$, we have:

$$
\begin{aligned}
C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},t)&=C(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}},t)+\frac{(q-1)c_f}{t}+c_f\left(P_{neg}(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}},t)-P_{neg}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},t)\right)\\
&<I_1+\frac{(q-1)c_f}{t}+c_f\left(P_{neg}(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}},t)-P_{neg}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},t)\right)\quad\text{(pooling is optimal for n-plex)}\\
&=I_q-(q-1)c_f+\frac{(q-1)c_f}{t}+c_f\left(P_{neg}(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}},t)-P_{neg}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},t)\right)\\
&=I_q+c_f\left(\underbrace{P_{neg}(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}},t)-P_{neg}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},t)}_{\Delta(q,t)}-\underbrace{(q-1)\left(1-\frac{1}{t}\right)}_{h(q,t)}\right).
\end{aligned}
$$

Thus, to prove that $C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, t) < I_q$, it suffices to show that, for $t \geq 2 : C(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}}, t) < I_1$,

$$\Delta(q,t) < h(q,t), \ \forall q \geq 2. \tag{22}$$

We do this by showing that the maximum of the LHS is strictly less than the minimum of the RHS of (22) for all $q \geq 2$ and any $t \geq 2 : C(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}}, t) < I_1$. The function $h(q,t)$ is strictly increasing in each of q and t. There are two cases for q:

Case 1 ($q \geq 3$). For any $q \geq 3$ and $t \geq 2$, $h(q,t) = (q-1)(1-1/t) \geq (3-1)(1-1/2) = 1$. Since $\Delta(q,t)$ is a difference of probabilities, $\Delta(q,t) < 1 \leq h(q,t)$, and the inequality holds for all $q \geq 3$.

Case 2 ($q = 2$).

$$\min_{t\geq 2} h(2,t) = (2-1)\left(1 - \frac{1}{2}\right) = 0.5. \tag{23}$$

Therefore, it is sufficient to show that $\max_{t\geq 2} \Delta(2,t) < 0.5$.

We have that $1 - \pi(\mathcal{S}) \geq p_0$, for any $\mathcal{S} \subseteq \mathcal{N}$. Therefore, by (21), for $q = 2$:

$$P_{neg}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}(q=2), t) \geq p_0 \cdot \prod_{k=1}^{2} p_0^{t-1} = p_0 \cdot p_0^{2(t-1)} = p_0^{2t-1}.$$

Thus, by (21), for any $p_0 \in (0,1)$, $\Delta(2,t) \leq p_0^t - p_0^{2t-1} < p_0^t - p_0^{2t}$, where the last inequality follows because $p_0 \in (0,1)$. Letting $u = p_0^t$ (where $u \in (0,1)$), the upper bound becomes $u - u^2$, and its maximum value occurs at $u = 0.5$ and equals 0.25. Therefore, for any $t \geq 2$:

$$\Delta(2,t) < 0.25. \tag{24}$$

Consequently, by Eqs. (23) and (24), $\Delta(2,t) < 0.25 < 0.5 \leq h(2,t)$, for any $t \geq 2$.

Thus, for all $q \geq 2$, we have shown that if $C(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}}, t) < I_1$ for some $t \geq 2$, then $C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, t) < I_q$, that is, if adaptive pooling is optimal for the n-plex, then adaptive pooling of at least one assay is also optimal for any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$, that is, $\boldsymbol{t}^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \neq \mathbf{1}$.

Part 2. By Definition 2, the given conditions, $\pi_i \geq \underline{p}(\{i\}), \ \forall i \in \mathcal{N}$, imply:

$$\begin{aligned}\pi_i \geq \underline{p}(\{i\}) &= \max_{t'\geq 2} p^*(\{i\}, t') \\ &\geq p^*(\{i\}, t) = \left(1 - \left((\rho(\{i\}, t))^+\right)^{1/t}\right)^+, \ \forall t \geq 2, \forall i \in \mathcal{N}.\end{aligned} \tag{25}$$

For any $i \in \mathcal{N}$, if $\exists t \geq 2$ with $\rho(\{i\}, t) \leq 0$, then $p^*(\{i\}, t) = 1 \Rightarrow \underline{p}(\{i\}) = 1$. Consequently, $\pi_i \geq \underline{p}(\{i\}) = 1$ cannot be satisfied (as $\pi_i \leq \pi(\mathcal{N}) < 1$ since $p_0 > 0$). Therefore, it must be true that $\rho(\{i\}, t) > 0, \forall t \geq 2, i \in \mathcal{N}$, which implies:

$$p^*(\{i\}, t) = \left(1 - (\rho(\{i\}, t))^{1/t}\right)^+, \forall t \geq 2, i \in \mathcal{N}. \tag{26}$$

By (25) and (26), and Definition 2, for any $i \in \mathcal{N}$,

$$\begin{aligned}&\pi_i \geq \underline{p}(\{i\}) \Rightarrow \pi_i \geq \left(1 - \left(\rho(\{i\}, t)\right)^{1/t}\right)^+ \geq 1 - \left(\rho(\{i\}, t)\right)^{1/t}, \quad \forall t \geq 2 \\ &\Rightarrow (1-\pi_i)^t \leq \rho(\{i\}, t), \quad \forall t \geq 2 \\ &\iff (1-\pi_i)^t (1 + \frac{c_v}{c_f}) \leq \frac{1}{t}\left(1 + \frac{c_v}{c_f}\right), \forall t \geq 2 \iff (1-\pi_i)^t \leq \frac{1}{t}, \quad \forall t \geq 2.\end{aligned} \tag{27}$$

By Theorem 2 and Definition 2, pooling is optimal for the n-plex iff $\pi(\mathcal{N}) < \underline{p}(\mathcal{N}) = \max_{t'\geq 2} p^*(\mathcal{N}, t') = p^*(\mathcal{N}, t)$ for some $t \geq 2$, equivalently, $\exists t \geq 2$ such that $\pi(\mathcal{N}) < p^*(\mathcal{N}, t) = \left(1 - \left((\rho(\mathcal{N}, t))^+\right)^{1/t}\right)^+$, where $\rho(\mathcal{N}, t) < 1$ (otherwise, $\pi(\mathcal{N}) < p^*(\mathcal{N}, t) = 0$, which cannot be satisfied). Further, by Definition 2 and (27),

$$\rho(\mathcal{N}, t) = \frac{1}{t}\left(1 + \frac{nc_v}{c_f}\right) - \frac{c_v}{c_f} \sum_{i\in\mathcal{N}} (1-\pi_i)^t \geq \frac{1}{t}\left(1 + \frac{nc_v}{c_f}\right) - \frac{nc_v}{c_f}\frac{1}{t} = \frac{1}{t} > 0.$$

Because $0 < \rho(\mathcal{N},t) < 1$, the adaptive pooling condition for the n-plex (Theorem 2) reduces to:

$$\pi(\mathcal{N}) < 1 - \left(\rho(\mathcal{N},t)\right)^{1/t} \iff (1-\pi(\mathcal{N}))^t + \frac{c_v}{c_f}\sum_{i\in\mathcal{N}}\left[(1-\pi_i)^t - \frac{1}{t}\right] > \frac{1}{t}. \tag{28}$$

However, because $\pi(\mathcal{N}) \geq \pi_i,\ \forall i \in \mathcal{N}$, we also have $(1-\pi(\mathcal{N}))^t \leq (1-\pi_i)^t \leq 1/t$ (by (27)). Therefore,

$$(1-\pi(\mathcal{N}))^t + \frac{c_v}{c_f}\sum_{i\in\mathcal{N}}\underbrace{\left[(1-\pi_i)^t - \frac{1}{t}\right]}_{\leq 0} \leq (1-\pi(\mathcal{N}))^t \leq \frac{1}{t},$$

contradicting (28). Hence, by Theorem 2, adaptive pooling cannot be optimal for the n-plex.
Part 3. Since $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$ (the given condition) and $\pi(\mathcal{S}) \leq \pi(\mathcal{N}),\ \forall \mathcal{S} \subseteq \mathcal{N}$, by Definition 2 $\pi(\mathcal{S}) \leq \pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi}) \leq \underline{p}(\mathcal{S}),\ \forall \mathcal{S} \subseteq \mathcal{N}$. Thus, $\pi(\mathcal{S}) < \underline{p}(\mathcal{S}), \forall \mathcal{S} \subseteq \mathcal{N}$, and the result follows by Theorem 2. $\square$

Proof of Proposition 2 *Part 1.* The result follows because for a fixed marginal vector $\boldsymbol{\pi}$, the adaptive pooling threshold $\underline{p}(\mathcal{S})$ is independent of the joint vector $\boldsymbol{p}$ (Definition 2), and $\pi(\mathcal{S}(\boldsymbol{p})) \leq \pi(\mathcal{S}(\boldsymbol{p}^{\text{nc}}(\boldsymbol{\pi}))),\ \forall \boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$. Consequently, if $\pi(\mathcal{S}(\boldsymbol{p}^{\text{nc}}(\boldsymbol{\pi}))) < \underline{p}(\mathcal{S})$, then $\pi(\mathcal{S}(\boldsymbol{p})) < \underline{p}(\mathcal{S}),\ \forall \boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$.
Part 2. Let $t \geq 2$ be fixed. By Definition 2, maximizing $\underline{p}(\mathcal{S})$ is equivalent to maximizing $p^*(\mathcal{S},t)$, which in turn is equivalent to minimizing $\rho(\mathcal{S},t) = \frac{1}{t}\left(1 + \frac{sc_v}{c_f}\right) - \frac{c_v}{c_f}\sum_{i\in\mathcal{S}}(1-\pi_i)^t$. Let $\boldsymbol{\pi}_{(\mathcal{S})} := (\pi_i)_{i\in\mathcal{S}}, \mathcal{S} \subseteq \mathcal{N}$. Thus, for a given $\sum_{i\in\mathcal{S}}\pi_i = \Pi$, we wish to maximize the summation term $F(\boldsymbol{\pi}_{(\mathcal{S})}) := \sum_{i\in\mathcal{S}}(1-\pi_i)^t$ over the convex polytope defined by $\sum_{i\in\mathcal{S}}\pi_i = \Pi$ and $\pi_i \geq \epsilon,\ \forall i \in \mathcal{S}$ (reflecting the distinct disease requirement, $\pi_i \geq p_i \geq \epsilon,\ \ \forall i \in \mathcal{N}$). For any $t \geq 2$, $g(\pi) := (1-\pi)^t$ is a strictly convex decreasing function of π on $(0,1)$; hence $F(\boldsymbol{\pi}_{(\mathcal{S})})$ is a strictly convex decreasing function, as it is the sum of strictly convex decreasing functions. The maximum of a strictly convex function over a convex polytope is achieved at an extreme point of the feasible region. In this symmetric setting, the extreme points correspond to marginal vectors where exactly one component is maximized (taking value $\Pi - (s-1)\epsilon$) and the others are minimized (taking value ϵ). $\square$

B.5. Proofs for §3.3.1

To simplify the exposition, we omit the superscripts $(c_v = 0)$ and $(c_f = 0)$ as well as the arguments in parentheses when the context is clear.

B.5.1. Supporting Results

Corollary B.2 (Optimal n-plex pool size for $c_v = 0$). *Consider that $c_v = 0$ and $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$. For $\boldsymbol{\mathcal{S}}^{n\text{-}plex}$, the optimal adaptive pool size equals the Dorfman pool size, that is, $t^{*(c_v=0)}(\mathcal{N}) = t^{Dorf}(\mathcal{N})$, which satisfies the FOC. In addition,*

$$C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{n\text{-}plex}, t^{*(c_v=0)}(\mathcal{N})) = C^{Dorf}(\boldsymbol{\mathcal{S}}^{n\text{-}plex}, t^{Dorf}(\mathcal{N})) < c_f.$$

Proof Let $c_v = 0$ and $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$. By Theorems 1 and 2:

$$C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{n\text{-plex}}, t(\mathcal{N})) = c_f \cdot \left(\frac{1}{t(\mathcal{N})} + 1 - (p_0)^{t(\mathcal{N})}\right) = C^{\text{Dorf}(c_v=0)}(\boldsymbol{\mathcal{S}}^{n\text{-plex}}, t(\mathcal{N})).$$

Further, $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$, thus $\pi(\mathcal{N}) < \underline{p}^{\text{Dorf}}$ because $\underline{p}^{\text{Dorf}} \leq \underline{p}_{\min}(\boldsymbol{\pi})$ (Theorem 2). Hence, the result, $t^{*(c_v=0)}(\mathcal{N}) = t^{\text{Dorf}}(\mathcal{N})$, readily follows from Proposition B.1, which satisfies the FOC. Finally, by Proposition B.1, $C^{\text{Dorf}}(\boldsymbol{\mathcal{S}}^{n\text{-plex}}, t^{\text{Dorf}}(\mathcal{N})) < c_f$, hence $C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{n\text{-plex}}, t^{*(c_v=0)}(\mathcal{N})) < c_f$. $\square$

We use the following definitions in the subsequent results. For any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$,

$$\beta_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) := -\frac{1}{2} \cdot \left(\frac{\gamma(\mathcal{S}_k)}{p_0 \cdot \prod_{\ell \in [q] \setminus \{k\}} (\gamma(\mathcal{S}_\ell))^{t_\ell - 1}} \cdot \ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2}, \ \forall \boldsymbol{t}_{-k}, k \in [q], \tag{29}$$

$$\tilde{\beta}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) := -\frac{1}{2} \cdot \Big(\sum_{\ell \in [q]} \alpha(\mathcal{S}_\ell)\Big) \cdot \left(\frac{\prod_{\ell \in [q]} \gamma(\mathcal{S}_\ell)}{p_0}\right)^{1/2}. \tag{30}$$

Lemma B.6 (Local minimum and maximum pool sizes for $c_v = 0$). *Consider that $c_v = 0$. For any q-partition $\boldsymbol{\mathcal{S}}^{Portfolio}$, consider assay $k \in [q]$ and any given $\boldsymbol{t}_{-k}$. We have:*

(i) $\beta_k(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k}) \le -1/e \Rightarrow C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k})$ *is strictly decreasing,* $\forall t_k > 0$.

(ii) $\beta_k(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k}) > -1/e \Rightarrow \exists$ *exactly two FOC solutions satisfying* $\frac{\partial C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k})}{\partial t_k} = 0$:

$$\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k}) = \frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot W_0(\beta_k(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k})),$$
$$\bar{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k}) = \frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot W_{-1}(\beta_k(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k})),$$

where $\underline{t}_k^{(c_v=0)}$ and $\bar{t}_k^{(c_v=0)}$ correspond to a local minimum and a local maximum, resp., and $\underline{t}_k^{(c_v=0)} < \bar{t}_k^{(c_v=0)}$. Further, $C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k})$ is decreasing in $t_k \in (0, \underline{t}_k^{(c_v=0)}) \cup (\bar{t}_k^{(c_v=0)}, \infty)$, and increasing in $t_k \in (\underline{t}_k^{(c_v=0)}, \bar{t}_k^{(c_v=0)})$.

In both cases, $\lim_{t_k \to \infty} C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k}) = c_f \cdot \Big(\big(\sum_{\ell \in [q] \setminus \{k\}} \frac{1}{t_\ell}\big) + 1 \Big) \ge c_f$.

Proof Consider any q-partition $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$. For $c_v = 0$, the cost function (5) (Theorem 1) is:

$$C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}) = c_f \cdot \Big(\big(\sum_{k \in [q]} \frac{1}{t_k}\big) + 1 - p_0 \cdot \prod_{k \in [q]} \big(\gamma(\mathcal{S}_k)\big)^{t_k - 1} \Big), \text{ where}$$

$$\frac{\partial C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})}{\partial t_k} = c_f \cdot \Big(-\frac{1}{t_k^2} - p_0 \cdot \ln(\gamma(\mathcal{S}_k)) \cdot (\gamma(\mathcal{S}_k))^{t_k - 1} \cdot \prod_{\ell \in [q] \setminus \{k\}} (\gamma(\mathcal{S}_\ell))^{t_\ell - 1} \Big), k \in [q].$$

First, we study the asymptotic behavior of the cost function and its derivative:

$$\begin{aligned}
&\lim_{t_k \to 0^+} C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) = \infty, \ \lim_{t_k \to \infty} C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) = c_f \cdot \Big(\big(\sum_{\ell \in [q] \setminus \{k\}} \frac{1}{t_\ell}\big) + 1 \Big) \ge c_f, \\
&\lim_{t_k \to 0^+} \frac{\partial C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})}{\partial t_k} = -\infty, \ \lim_{t_k \to \infty} \frac{\partial C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})}{\partial t_k} = 0^-, \\
&\frac{\partial C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})}{\partial t_k} = 0 \ \Rightarrow \ -t_k^2 \cdot (\gamma(\mathcal{S}_k))^{t_k} \cdot \ln(\gamma(\mathcal{S}_k)) = \frac{\gamma(\mathcal{S}_k)}{p_0 \cdot \prod_{\ell \in [q] \setminus \{k\}} (\gamma(\mathcal{S}_\ell))^{t_\ell - 1}}, \ \forall k \in [q].
\end{aligned} \tag{31}$$

After some algebraic manipulations (i.e., for each side, multiplying by $-\ln(\gamma(\mathcal{S}_k))$, taking the square root, dividing by 2), and using the equivalence, $(\gamma(\mathcal{S}_k))^{t_k/2} = e^{\frac{t_k \cdot \ln(\gamma(\mathcal{S}_k))}{2}}$, we obtain:

$$\frac{t_k \cdot \ln(\gamma(\mathcal{S}_k))}{2} \cdot e^{\frac{t_k \cdot \ln(\gamma(\mathcal{S}_k))}{2}} = \pm \frac{1}{2} \cdot \left(\frac{\gamma(\mathcal{S}_k)}{p_0 \cdot \prod_{\ell \in [q] \setminus \{k\}} (\gamma(\mathcal{S}_\ell))^{t_\ell - 1}} \cdot \ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2}, \ \forall k \in [q].$$

We drop the arguments of β_k to simplify the subsequent notation. Because the LHS is negative, it is sufficient to consider the solution with $-1/2$ coefficient, that is, the FOC becomes $\frac{t_k \cdot \ln(\gamma(\mathcal{S}_k))}{2} \cdot e^{\frac{t_k \cdot \ln(\gamma(\mathcal{S}_k))}{2}} = \beta_k$. Then, if it exists, the solution can be obtained from the Lambert-W function definition of $x \cdot e^x = a \ \Rightarrow$

$x = W(a)$. There are two possible cases:
Case (i). $\beta_k \leq -1/e$: The Lambert-W function is not defined, and no finite FOC solution exists for t_k. Then, due to (31), $C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})$ must decrease, $\forall t_k > 0$, establishing part (i).
Case (ii). Otherwise ($\beta_k > -1/e$): By Property B.2, FOC has two real solutions given by:

$$\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) = \frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot W_0(\beta_k) \quad \text{and} \quad \bar{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) = \frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot W_{-1}(\beta_k),$$

where $\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) < \bar{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})$.

Due to the asymptotic behavior in (31), the cost function is decreasing for sufficiently small and large t_k values. Since there are exactly two FOCs, the cost function must be,

- decreasing in $t_k \in (0, \underline{t}_k^{(c_v=0)})$, equivalently, $\frac{\partial C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})}{\partial t_k} < 0$;
- increasing in $t_k \in (\underline{t}_k^{(c_v=0)}, \bar{t}_k^{(c_v=0)})$, equivalently, $\frac{\partial C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})}{\partial t_k} > 0$; and
- decreasing in $t_k \in (\bar{t}_k^{(c_v=0)}, \infty)$, equivalently, $\frac{\partial C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})}{\partial t_k} < 0$.

Therefore, $\underline{t}_k^{(c_v=0)}$ and $\bar{t}_k^{(c_v=0)}$ resp. correspond to the local minimum and local maximum pool sizes of the cost function. This completes the proof. □

Proposition B.2 (Optimal pool sizes for $c_v = 0$). *Consider that $c_v = 0$ and $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$. For any q-partition $\boldsymbol{\mathcal{S}}^{Portfolio}$, the FOC pool size solutions exist for all assays only if $\tilde{\beta}(\boldsymbol{\mathcal{S}}^{Portfolio}) > -1/e$. Regarding the optimal pool size vector $\boldsymbol{t}^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio})$:*

1. *Either $t_k^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio})$, $\forall k \in [q]$ satisfies the FOC, yielding*

$$t_k^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}) = \frac{\tilde{t}(\boldsymbol{\mathcal{S}}^{Portfolio})}{\alpha(\mathcal{S}_k)}, \ \forall k \in [q], \ \textit{where } \tilde{t}(\boldsymbol{\mathcal{S}}^{Portfolio}) := \frac{-2}{\sum_{\ell \in [q]} \alpha(\mathcal{S}_\ell)} \cdot W_0(\tilde{\beta}(\boldsymbol{\mathcal{S}}^{Portfolio})).$$

   Hence, $\pi(\mathcal{S}_k) > \pi(\mathcal{S}_\ell) \Rightarrow t_k^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}) < t_\ell^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio})$ for any $k, \ell \in [q]$.

2. *Or $t_k^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}) \to \boldsymbol{\infty}$, $\forall k \in [q]$, with $\lim_{\boldsymbol{t}^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}) \to \boldsymbol{\infty}} C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio})) = c_f$.*

Proof Let $c_v = 0$. Consider any q-partition $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$. As $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$, $\boldsymbol{t}^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \neq \boldsymbol{1}$ (Proposition 1). Thus, each $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$, $\forall k \in [q]$ must either correspond to the boundary solution $t_{\texttt{max}}(\to \infty)$ (part 2 of the proposition) or satisfy the FOC (part 1 of the proposition). Then, it suffices to consider the following cost function ((5) in Theorem 1):

$$C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}) = c_f \cdot \Big(\big(\sum_{k \in [q]} \frac{1}{t_k} \big) + 1 - p_0 \cdot \prod_{k \in [q]} \big(p_0(\mathcal{S}_k)\big)^{t_k - 1} \Big).$$

First we show that if $\exists k \in [q]$ such that $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \to \infty$, then $t_\ell^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \to \infty$, $\forall \ell \in [q]$. To this end, assume that $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \to \infty$ for some $k \in [q]$, with the cost function reducing to,

$$\lim_{t_k^* \to \infty} C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, t_k^*, \boldsymbol{t}_{-k}) = c_f \cdot \Big(\big(\sum_{\ell \in [q] \setminus \{k\}} \frac{1}{t_\ell} \big) + 1 \Big),$$

which is separable by assay, yielding $t_\ell^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, t_k^* \to \infty) \to \infty$, $\forall \ell \in [q] \setminus \{k\}$. Hence it follows that $\lim_{\boldsymbol{t}^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \to \boldsymbol{\infty}} C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})) = c_f$, establishing part 2.

Next we consider the remaining case, i.e., $\nexists k \in [q]$ such that $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \to \infty$. Then, by Theorem 2 all pool sizes must correspond to the FOC, $\partial C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}))/\partial t_k = 0$, $\forall k \in [q]$, which, from Lemma B.6, implies:

$$-(t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}))^2 \cdot \ln(\gamma(\mathcal{S}_k)) = \frac{1}{p_0 \cdot \prod_{\ell \in [q]} (\gamma(\mathcal{S}_\ell))^{t_\ell^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) - 1}}, \ \forall k \in [q] \tag{32}$$

$$\Rightarrow -(t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}))^2 \cdot \ln(\gamma(\mathcal{S}_k)) = -(t_\ell^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}))^2 \cdot \ln(\gamma(\mathcal{S}_\ell)), \ \forall \ell, k \in [q]. \tag{33}$$

Note that (33) holds since the RHS of (32), which includes all assays in the product term, remains the same for all assay indices. Then, it readily follows from (33) that if $\pi(\mathcal{S}_k) > \pi(\mathcal{S}_\ell)$ (equivalently, $\gamma(\mathcal{S}_k) < \gamma(\mathcal{S}_\ell)$), then $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) < t_\ell^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$, the last result in part 1.
Next, we characterize the optimal pool size vector that satisfies the FOC. Observe that:

$$(33) \Rightarrow (t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}))^2 \cdot \ln(\frac{1}{\gamma(\mathcal{S}_k)}) = (t_\ell^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}))^2 \cdot \ln(\frac{1}{\gamma(\mathcal{S}_\ell)}), \ \forall \ell, k \in [q].$$

Taking the square root of both sides and substituting the definition of $\alpha(\mathcal{S}_k) = \Big(\ln(\frac{1}{\gamma(\mathcal{S}_k)})\Big)^{1/2} (>0)$, we get $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \cdot \alpha(\mathcal{S}_k) = t_\ell^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \cdot \alpha(\mathcal{S}_\ell), \ \forall \ell, k \in [q]$. Letting,

$$\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) := t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \cdot \alpha(\mathcal{S}_k), \ \forall k \in [q], \tag{34}$$

$$\prod_{\ell \in [q]} (\gamma(\mathcal{S}_\ell))^{t_\ell^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})-1} = \frac{\prod_{\ell \in [q]} (\gamma(\mathcal{S}_\ell))^{t_\ell^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})}}{\prod_{\ell \in [q]} \gamma(\mathcal{S}_\ell)} = \frac{\prod_{\ell \in [q]} \Big(\gamma(\mathcal{S}_\ell))^{(\alpha(\mathcal{S}_\ell))^{-1}}\Big)^{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})}}{\prod_{\ell \in [q]} \gamma(\mathcal{S}_\ell)}. \tag{35}$$

Then, by (32), (34), and (35),

$$(\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}))^2 = \frac{\prod_{\ell \in [q]} \gamma(\mathcal{S}_\ell)}{p_0 \cdot \prod_{\ell \in [q]} \Big(\gamma(\mathcal{S}_\ell))^{(\alpha(\mathcal{S}_\ell))^{-1}}\Big)^{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})}}.$$

To simplify the subsequent notation, let $K := \prod_{\ell \in [q]} (\gamma(\mathcal{S}_\ell))^{(\alpha(\mathcal{S}_\ell))^{-1}}$, hence we have:

$$(\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}))^2 \cdot K^{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})} = \frac{\prod_{\ell \in [q]} \gamma(\mathcal{S}_\ell)}{p_0}.$$

After some algebraic manipulations (i.e., for each side, multiplying by $\ln^2(K)$, taking the square root, dividing by 2), and using the equivalence, $K^{\frac{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})}{2}} = e^{\frac{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \cdot \ln(K)}{2}}$:

$$\frac{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \cdot \ln(K)}{2} \cdot e^{\frac{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \cdot \ln(K)}{2}} = \pm \frac{1}{2} \cdot \left(\frac{\prod_{\ell \in [q]} \gamma(\mathcal{S}_\ell)}{p_0} \cdot \ln^2(K) \right)^{1/2}.$$

Because the LHS is negative, it is sufficient to consider the solution with $-1/2$ coefficient on the RHS. Then, if it exists, the FOC solution can be obtained from the Lambert-W function definition of $x \cdot e^x = a \ \Rightarrow \ x = W(a)$. Regarding the existence of the FOC solution, note that:

$$\tilde{\beta}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) := -\frac{1}{2} \cdot \Big(\sum_{\ell \in [q]} \alpha(\mathcal{S}_\ell) \Big) \cdot \left(\frac{\prod_{\ell \in [q]} \gamma(\mathcal{S}_\ell)}{p_0} \right)^{1/2},$$

is equivalent to the RHS since $\ln(K) = -\sum_{\ell \in [q]} \Big| \sqrt{-\ln(\gamma(\mathcal{S}_\ell))} \Big| = -\sum_{\ell \in [q]} \alpha(\mathcal{S}_\ell)$.

Note that $\tilde{\beta}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \le 0$. If $\tilde{\beta}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \le -1/e$, the Lambert-W function is not defined, and there would not be any finite solutions for $\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ (equivalently, a solution to FOC does not exist), whereas if $\tilde{\beta}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) > -1/e$, then exactly two FOC solutions exist, proving the existence condition in the proposition. In the latter case, one of the FOC solutions corresponds to $\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$: $\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) = \frac{2}{\ln(K)} \cdot W_0(\tilde{\beta}) = \frac{-2}{\sum_{\ell \in [q]} \alpha(\mathcal{S}_\ell)} \cdot W_0\left(\tilde{\beta}\right)$. Using this, the optimal pool sizes obtained from (34) match the optimal pool sizes provided in part 1. □

Lemma B.7 (Merged assay prevalence properties). *Consider any pair of nonempty assays, $\mathcal{S}_k, \mathcal{S}_\ell \subseteq \mathcal{N}$, and let $\mathcal{S}_{k\ell} := \mathcal{S}_k \cup \mathcal{S}_\ell$. If $\pi(\mathcal{S}_{k\ell}) \in (0, 1-e^{-0.5}) \approx (0, 0.393)$, then:*

$$(i)\ \alpha(\mathcal{S}_{k\ell}) < \sum_{r\in\{k,\ell\}} \alpha(\mathcal{S}_r) \quad \textit{and} \quad (ii)\ \prod_{r\in\{k,\ell\}} \left(\gamma(\mathcal{S}_r)\right)^{\frac{1}{\alpha(\mathcal{S}_r)}} < \left(\gamma(\mathcal{S}_{k\ell})\right)^{\frac{1}{\alpha(\mathcal{S}_{k\ell})}}.$$

<u>Proof</u> We use the notation $\alpha(\mathcal{S})$ and $\alpha(\gamma(\mathcal{S}))$ interchangeably. Observe that the function $\alpha(\mathcal{S}) = (\ln(\frac{1}{\gamma(\mathcal{S})}))^{1/2} > 0$ is:

- decreasing in $\gamma(\mathcal{S}) \in (0,1)$, since $\dfrac{\partial \alpha(\mathcal{S})}{\partial \gamma(\mathcal{S})} = -\dfrac{1}{2\gamma(\mathcal{S})[-\ln(\gamma(\mathcal{S}))]^{1/2}} < 0$;
- concave decreasing in $\gamma(\mathcal{S}) \in (e^{-0.5}, 1)$, since $\dfrac{\partial^2 \alpha(\mathcal{S})}{\partial (\gamma(\mathcal{S}))^2} = \dfrac{-[2\ln(\gamma(\mathcal{S}))+1]}{4(\gamma(\mathcal{S}))^2 \cdot [-\ln(\gamma(\mathcal{S}))]^{3/2}} < 0.$

Then, since $\mathcal{S}_{k\ell} = \mathcal{S}_k \cup \mathcal{S}_\ell$, and $\alpha(\mathcal{S})$ is decreasing in $\gamma(\mathcal{S})$, we have, for all $r \in \{k, \ell\}$,

$$\pi(\mathcal{S}_{k\ell}) > \pi(\mathcal{S}_r) \ \Rightarrow\ \gamma(\mathcal{S}_{k\ell}) < \gamma(\mathcal{S}_r) \ \Rightarrow\ \alpha(\mathcal{S}_{k\ell}) > \alpha(\mathcal{S}_r) \ \Rightarrow\ \frac{1}{\alpha(\mathcal{S}_{k\ell})} < \frac{1}{\alpha(\mathcal{S}_r)}. \tag{36}$$

Case 1. Suppose $\gamma(\mathcal{S}_{k\ell}) \geq \prod_{r\in\{k,\ell\}} \gamma(\mathcal{S}_r)$. We have:

$$\begin{aligned}\ln(\gamma(\mathcal{S}_{k\ell})) \geq \ln\Big(\prod_{r\in\{k,\ell\}} \gamma(\mathcal{S}_r)\Big) \ \Rightarrow\ -\ln(\gamma(\mathcal{S}_{k\ell})) \leq -\ln\Big(\prod_{r\in\{k,\ell\}} \gamma(\mathcal{S}_r)\Big) = -\sum_{r\in\{k,\ell\}} \ln(\gamma(\mathcal{S}_r)) \\ \Rightarrow \ln\Big(\frac{1}{\gamma(\mathcal{S}_{k\ell})}\Big) \leq \sum_{r\in\{k,\ell\}} \ln\Big(\frac{1}{\gamma(\mathcal{S}_r)}\Big),\end{aligned}$$

which yields, after taking the square root of both sides (and using the positive roots),

$$\alpha(\mathcal{S}_{k\ell}) \leq \Big[\sum_{r\in\{k,\ell\}} \ln\Big(\frac{1}{\gamma(\mathcal{S}_r)}\Big)\Big]^{1/2} < \sum_{r\in\{k,\ell\}} \alpha(\mathcal{S}_r),$$

which holds since the square root function is strictly concave on non-negative reals. Thus, (i) holds.

For $r \in \{k, \ell\}$, because $0 < \gamma(\mathcal{S}_r) < 1$, we have $\big(\gamma(\mathcal{S}_r)\big)^{\frac{1}{\alpha(\mathcal{S}_r)}} < \big(\gamma(\mathcal{S}_r)\big)^{\frac{1}{\alpha(\mathcal{S}_{k\ell})}}$ by (36). This implies the first inequality in the following:

$$\prod_{r\in\{k,\ell\}} \big(\gamma(\mathcal{S}_r)\big)^{\frac{1}{\alpha(\mathcal{S}_r)}} < \Big(\prod_{r\in\{k,\ell\}} \big(\gamma(\mathcal{S}_r)\big)\Big)^{\frac{1}{\alpha(\mathcal{S}_{k\ell})}} \leq \big(\gamma(\mathcal{S}_{k\ell})\big)^{\frac{1}{\alpha(\mathcal{S}_{k\ell})}},$$

whereas the second inequality follows from the assumption of Case 1. Hence, (ii) holds.
Case 2. Suppose $\gamma(\mathcal{S}_{k\ell}) < \prod_{r\in\{k,\ell\}} \gamma(\mathcal{S}_r)$. Based on (36), we represent $\gamma(\mathcal{S}_k) = \gamma(\mathcal{S}_{k\ell}) + \delta_k$ and $\gamma(\mathcal{S}_\ell) = \gamma(\mathcal{S}_{k\ell}) + \delta_\ell$ for some $\delta_k, \delta_\ell > 0$. As we show next, this also implies $\delta_k + \delta_\ell + \gamma(\mathcal{S}_{k\ell}) \leq 1$:

$$\begin{aligned}\delta_k + \delta_\ell + \gamma(\mathcal{S}_{k\ell}) &= \gamma(\mathcal{S}_k) - \cancel{\gamma(\mathcal{S}_{k\ell})} + \gamma(\mathcal{S}_\ell) - \gamma(\mathcal{S}_{k\ell}) + \cancel{\gamma(\mathcal{S}_{k\ell})} \\ &= (1 - \pi(\mathcal{S}_k)) + (\cancel{1} - \pi(\mathcal{S}_\ell)) - (\cancel{1} - \pi(\mathcal{S}_{k\ell})) = 1 - [\pi(\mathcal{S}_k) + \pi(\mathcal{S}_\ell) - \pi(\mathcal{S}_{k\ell})] \\ &\overset{(2.1.3)}{=} 1 - \Big[\sum_{c\in\mathcal{I}^{\mathtt{cml}}(\mathcal{S}_k)} \big(p_c + \sum_{c'\in\mathcal{I}^{\mathtt{cml}}(\mathcal{N}\setminus\mathcal{S}_k)} p_{c\oplus c'}\big) + \sum_{c\in\mathcal{I}^{\mathtt{cml}}(\mathcal{S}_\ell)} \big(p_c + \sum_{c'\in\mathcal{I}^{\mathtt{cml}}(\mathcal{N}\setminus\mathcal{S}_\ell)} p_{c\oplus c'}\big) - \sum_{c\in\mathcal{I}^{\mathtt{cml}}(\mathcal{S}_{k\ell})} \big(p_c + \sum_{c'\in\mathcal{I}^{\mathtt{cml}}(\mathcal{N}\setminus\mathcal{S}_{k\ell})} p_{c\oplus c'}\big)\Big] \\ &= 1 - \sum_{c_k\in\mathcal{I}^{\mathtt{cml}}(\mathcal{S}_k)} \ \sum_{c_\ell\in\mathcal{I}^{\mathtt{cml}}(\mathcal{S}_\ell)} \ \sum_{c'\in\mathcal{I}^{\mathtt{cml}}(\mathcal{N}\setminus\mathcal{S}_{k\ell})\cup\{0\}} p_{c_k\oplus c_\ell\oplus c'} \leq 1.\end{aligned} \tag{37}$$

Under the given condition, $\pi(\mathcal{S}_{k\ell}) \in (0, 1-e^{-0.5})$, i.e, $\gamma(\mathcal{S}_{k\ell}) \in (e^{-0.5}, 1)$, thus, $\gamma(\mathcal{S}_k), \gamma(\mathcal{S}_\ell) \in (e^{-0.5}, 1)$ ((36)). To show $\alpha(\mathcal{S}_k) + \alpha(\mathcal{S}_\ell) - \alpha(\mathcal{S}_{k\ell}) > 0$, we analyze the optimization problem:

$$\begin{aligned} \min_{\delta_k, \delta_\ell} \; & \alpha(\delta_k + \gamma(\mathcal{S}_{k\ell})) + \alpha(\delta_\ell + \gamma(\mathcal{S}_{k\ell})) - \alpha(\gamma(\mathcal{S}_{k\ell})) \\ \text{subject to } & \epsilon \le \delta_k, \quad \epsilon \le \delta_\ell, \quad \delta_\ell + \delta_k \le 1 - \gamma(\mathcal{S}_{k\ell}), \end{aligned}$$

where $\epsilon > 0$ is sufficiently small to ensure $\gamma_l, \gamma_k > 0$, and the last constraint follows by (37). Because $\alpha(\cdot)$ is concave decreasing in $(e^{-0.5}, 1)$ and the sum of two concave decreasing functions is concave decreasing, the objective function is concave decreasing in $(e^{-0.5}, 1)$. Since the feasible set is a polytope (hence convex and compact), it has an extreme point that is a global minimum. Hence, there are two candidates for the optimal solution $(\delta_k^*, \delta_\ell^*)$: (ϵ, ϵ) and $(\epsilon, 1-\gamma(\mathcal{S}_{k\ell}) - \epsilon)$ (or its symmetric version). Comparing their objective values, and using the fact that $\alpha(\cdot)$ is decreasing, we find that the latter solution is optimal (as $\gamma(\mathcal{S}_{k\ell}) \le 1 - 2\epsilon$), yielding the minimum value,

$$\alpha(1-\epsilon) - \big[\alpha(\gamma(\mathcal{S}_{k\ell})) - \alpha(\epsilon + \gamma(\mathcal{S}_{k\ell}))\big] = \big[\alpha(1-\epsilon) - \alpha(1-\epsilon+\epsilon)\big] - \big[\alpha(\gamma(\mathcal{S}_{k\ell})) - \alpha(\gamma(\mathcal{S}_{k\ell}) + \epsilon)\big] > 0,$$

where the last inequality follows because $\alpha(\cdot)$ is a concave decreasing function, proving that $\alpha(\mathcal{S}_k) + \alpha(\mathcal{S}_\ell) - \alpha(\mathcal{S}_{k\ell}) > 0$ when $\gamma(\mathcal{S}_{k\ell}) \in (e^{-0.5}, 1]$.

To prove the second inequality given in (ii), we use the observation,

$$\ln\left(\gamma(\mathcal{S}_k)^{\frac{1}{\alpha(\mathcal{S}_k)}}\right) = \frac{1}{\alpha(\mathcal{S}_k)} \ln(\gamma(\mathcal{S}_k)) = \frac{1}{\sqrt{-\ln(\gamma(\mathcal{S}_k))}} \ln(\gamma(\mathcal{S}_k)) = -\sqrt{-\ln(\gamma(\mathcal{S}_k))} = -\alpha(\mathcal{S}_k),$$

along with the definition of $\alpha(\mathcal{S}) = \left(\ln(\frac{1}{\gamma(\mathcal{S})})\right)^{1/2}$. Then (ii) follows from (i) because:

$$\begin{aligned} \text{(i) } \alpha(\mathcal{S}_k) + \alpha(\mathcal{S}_\ell) > \alpha(\mathcal{S}_{k\ell}) \;&\Rightarrow\; -\alpha(\mathcal{S}_k) - \alpha(\mathcal{S}_\ell) < -\alpha(\mathcal{S}_{k\ell}) \\ &\Rightarrow \sum_{r \in \{k,\ell\}} \ln\left(\gamma(\mathcal{S}_r)^{\frac{1}{\alpha(\mathcal{S}_r)}}\right) < \ln\left(\gamma(\mathcal{S}_{k\ell})^{\frac{1}{\alpha(\mathcal{S}_{k\ell})}}\right) \quad \text{(using the observation)} \\ &\Rightarrow \prod_{r \in \{k,\ell\}} (\gamma(\mathcal{S}_r))^{\frac{1}{\alpha(\mathcal{S}_r)}} < (\gamma(\mathcal{S}_{k\ell}))^{\frac{1}{\alpha(\mathcal{S}_{k\ell})}} \qquad \text{(via exponentiation).} \quad \square \end{aligned}$$

B.5.2. Main Results

Proof of Theorem 3 *Part 1 (a).* Consider $c_v = 0$ and $\pi(\mathcal{N}) \le 1 - e^{-0.5}$. From (3):

$$C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})^{(c_v=0)} = c_f \cdot \Big(\Big[\sum_{k \in \mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})} \frac{1}{t_k} \Big] + 1 - \mathbb{P}\left(D_{\texttt{subject}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}) = 0\right) + n - n_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}) \Big),$$

where the first term represents the number of stage 1 pooled tests, the term $1 - \mathbb{P}\left(D_{\texttt{subject}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}) = 0\right)$ represents the expected number of stage 2 retests, and $n - n_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})\Big)$ represents the number of stage 1 individual tests, per subject. Thus, when $c_v = 0$, the minimization of the adaptive cost and per-subject expected number of tests coincide.

Part 1 (b). Consider $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$. We wish to show that, for any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$,

$$C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}}, t^{*(c_v=0)}(\mathcal{N})) \le C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})).$$

By Proposition B.2, when $c_v = 0$, the optimal pool size vector for any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$ is such that all pool sizes either satisfy the FOC or equal $t_{\texttt{max}}$ $(\to \infty)$. We show $\boldsymbol{\mathcal{S}}^{n\texttt{-plex}}$ optimality in both cases.

First, by Corollary B.2, for $\boldsymbol{\mathcal{S}}^{n\texttt{-plex}}$, the optimal pool size is $t^{*(c_v=0)}(\mathcal{N}) = t^{\texttt{Dorf}}(\mathcal{N})$ (satisfying the FOC) and the optimal cost satisfies,

$$C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{n\texttt{-plex}}, t^{*(c_v=0)}(\mathcal{N})) < c_f. \tag{38}$$

Suppose, to the contrary, that the optimal portfolio is not an n-plex, that is, $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}} = (\mathcal{S}_k)_{k\in[q]}$ for some $q \geq 2$. We first consider the case where all optimal pool sizes satisfy the FOC, for which a necessary (but not sufficient) condition is that, $\tilde{\beta}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) > -1/e$ (Proposition B.2), where $\tilde{\beta}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ is defined in (30). We construct a new portfolio by combining any two assays in the given portfolio, say assays $\mathcal{S}_1$ and $\mathcal{S}_2$, wlog, into $\mathcal{S}_{12} := \mathcal{S}_1 \cup \mathcal{S}_2$, while keeping assays $\mathcal{S}_3, \cdots, \mathcal{S}_q$ intact; we denote the new $(q-1)$-partition by $\hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}}$. We show that the cost of the new portfolio $\hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}}$ is lower than that of the given portfolio $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$, that is, merging any two assays in any multi-assay partition reduces the cost.
By the given condition, $\pi(\mathcal{S}_{k\ell}) \leq \pi(\mathcal{N}) \leq 1 - e^{-0.5}$, $\forall k, \ell \in [q]$. Hence, by Lemma B.7,

$$\alpha(\mathcal{S}_{12}) < \sum_{r=1}^{2} \alpha(\mathcal{S}_r) \quad \text{and} \quad \prod_{r=1}^{2} (\gamma(\mathcal{S}_r))^{\frac{1}{\alpha(\mathcal{S}_r)}} < (\gamma(\mathcal{S}_{12}))^{\frac{1}{\alpha(\mathcal{S}_{12})}}. \tag{39}$$

Substituting the optimal pool sizes (that correspond to the FOC) from Proposition B.2, namely $t_k^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) = \frac{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})}{\alpha(\mathcal{S}_k)}$, $\forall k \in [q]$, into the cost function $C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})^{(c_v=0)} = c_f \cdot \Big(\big(\sum_{k\in[q]} \frac{1}{t_k} \big) + 1 - p_0 \cdot \prod_{k\in[q]} \big(\gamma(\mathcal{S}_k)\big)^{t_k - 1} \Big)$ (Theorem 1), allows us to write the testing cost function parametrized by the single pool size $\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$:

$$\begin{aligned}
C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})) &= c_f \cdot \left[\frac{\sum_{k=1}^{q} \alpha(\mathcal{S}_k)}{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})} + 1 - p_0 \cdot \prod_{k=1}^{q} \big(\gamma(\mathcal{S}_k)\big)^{\frac{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})}{\alpha(\mathcal{S}_k)} - 1} \right] \\
&\overset{(39)}{>} c_f \cdot \left[\frac{\alpha(\mathcal{S}_{12}) + \sum_{k=3}^{q} \alpha(\mathcal{S}_k)}{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})} + 1 - p_0 \cdot \big(\gamma(\mathcal{S}_{12})\big)^{\frac{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})}{\alpha(\mathcal{S}_{12})} - 1} \cdot \prod_{k=3}^{q} \big(\gamma(\mathcal{S}_k)\big)^{\frac{\tilde{t}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})}{\alpha(\mathcal{S}_k)} - 1} \right] \\
&\geq C^{(c_v=0)}(\hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}}, \tilde{t}(\hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}})) \qquad \text{(by optimality of } \tilde{t}(\hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}}) \text{ for } \hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}}).
\end{aligned}$$

Proceeding in this way, i.e., merging assays and reducing the cost, we end up with n-plex as the optimal portfolio.
Next we consider the other case where $\boldsymbol{t}^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \to \boldsymbol{\infty}$. As shown in Proposition B.2,

$$\lim_{\boldsymbol{t}^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \to \boldsymbol{\infty}} C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}^{*(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})) = c_f.$$

By (38), $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$ with any $q \geq 2$ cannot be optimal, and the optimality of $\boldsymbol{\mathcal{S}}^{n\texttt{-plex}}$ follows.

Part 2 (a). Consider $c_f = 0$. By Theorem 1, (3) reduces to:

$$\begin{aligned}
C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})^{(c_f=0)} &= c_v \cdot \Big(\Big[\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})} \sum_{i\in\mathcal{S}_k} \frac{1}{w_i(\boldsymbol{t})} \Big] + \Big[n_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}) - \sum_{i\in\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})} (1-\pi_i)^{w_i(\boldsymbol{t})} \Big] + \sum_{k\in\mathcal{K}_{\texttt{ind}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})} s_k \Big) \\
&= c_v \cdot \Big(\Big[\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})} \sum_{i\in\mathcal{S}_k} \frac{1}{w_i(\boldsymbol{t})} \Big] + \Big[\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})} \big(s_k - \sum_{i\in\mathcal{S}_k} (1-\pi_i)^{w_i(\boldsymbol{t})} \big) \Big] + \sum_{k\in\mathcal{K}_{\texttt{ind}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})} s_k \Big) \\
&= c_v \cdot \Big(\Big[\sum_{k\in\mathcal{K}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})} \Big(s_k + \sum_{i\in\mathcal{S}_k} \big(\frac{1}{w_i(\boldsymbol{t})} - (1-\pi_i)^{w_i(\boldsymbol{t})} \big) \Big) \Big] + \sum_{k\in\mathcal{K}_{\texttt{ind}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})} s_k \Big),
\end{aligned}$$

where the last expression decomposes by assay (index k) for a given portfolio.

Part 2 (b). By Theorem 2, for any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$, $\mathcal{N}_{\texttt{pool}}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})) = \mathcal{N}$, hence the adaptive cost function (5) (Theorem 1) reduces to:

$$C^{(c_f=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}) = c_v \cdot \left(\sum_{i\in\mathcal{N}} \Big[\frac{1}{w_i(\boldsymbol{t})} + 1 - (\gamma_i)^{w_i(\boldsymbol{t})} \Big] \right) = c_v \cdot \left(\sum_{k\in[q]} \sum_{i\in\mathcal{S}_k} \Big[\frac{1}{t_k} + 1 - (\gamma_i)^{t_k} \Big] \right),$$

which is additively separable over assays. In the following, we start with an all-singleton portfolio $\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}$ with optimal pool sizes, then show that merging any two assays cannot reduce the cost.

Consider the all-singleton portfolio $\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}$, for which

$$t_i^{*(c_f=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}) = \arg\min_{t_i} \left\{ \frac{1}{t_i} + 1 - (\gamma_i)^{t_i} \right\} = t^{\texttt{Dorf}}(\{i\}),\ i \in \mathcal{N},$$

which follows because if $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$, then the optimal pool size is $t^{\texttt{Dorf}}(\{i\})$ (Corollary B.2).

We now merge any two singletons in this portfolio, say $\mathcal{S}_k = \{k\}$ and $\mathcal{S}_\ell = \{\ell\}, k, \ell \in \mathcal{N}, k \neq \ell$, into a new assay $\mathcal{S}_{k\ell} = \{k, \ell\}$, while keeping all other singletons ($\mathcal{S}_r = \{r\},\ \forall r \in \mathcal{N} \setminus \{k, \ell\}$) intact, yielding $\hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}}$. Because the pool size decision decomposes by assay, the optimal pool size vector for $\hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}}$ remains unchanged for all assays except for the new assay $\mathcal{S}_{k\ell}$, the cost of which is,

$$\min_{\hat{t}} \left\{ \sum_{r \in \{k,\ell\}} \left[\frac{1}{\hat{t}} + 1 - (\gamma_r)^{\hat{t}} \right] \right\} = \min_{t_k, t_\ell} \left\{ \sum_{r \in \{k,\ell\}} \left[\frac{1}{t_r} + 1 - (\gamma_r)^{t_r} \right] :\ t_k = t_\ell \right\} \geq \sum_{r \in \{k,\ell\}} \min_{t_r} \left\{ \frac{1}{t_r} + 1 - (\gamma_r)^{t_r} \right\}.$$

Consequently, it must be true that:

$$C(\hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}}, \boldsymbol{t}^{*(c_f=0)}(\hat{\boldsymbol{\mathcal{S}}}^{\texttt{Portfolio}})) \geq C(\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}, \boldsymbol{t}^{*(c_f=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Singletons}})) \geq 0.$$

Note that any portfolio can be obtained from $\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}$ by applying a finite sequence of assay merges, where a *merge* replaces two assays in a portfolio, $\mathcal{S}$ and $\mathcal{S}'$ with $\mathcal{S} \cap \mathcal{S}' = \emptyset$, with their union, $\mathcal{S} \cup \mathcal{S}'$. Next, we show via induction that no sequence of merges can yield a lower cost portfolio, hence $\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}$ is optimal.

Base Cases (0 and 1 merge): No merge is the case of $\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}$, and the case of one merge applied to $\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}$ does not result in any cost reduction, as shown above.

Inductive Hypothesis (m merges): Assume that for any portfolio $\boldsymbol{\mathcal{S}}_m^{\texttt{Portfolio}}$ obtained via m sequential merges starting from $\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}$, it is true that $C(\boldsymbol{\mathcal{S}}_m^{\texttt{Portfolio}}) \geq C(\boldsymbol{\mathcal{S}}^{\texttt{Singletons}})$.

Inductive Step ($m+1$ merges): Let $\boldsymbol{\mathcal{S}}_{m+1}^{\texttt{Portfolio}}$ be a portfolio formed by $(m+1)$ merges from singletons via the sequence $\boldsymbol{\mathcal{S}}^{\texttt{Singletons}} \xrightarrow{\text{merge } 1} \boldsymbol{\mathcal{S}}_1^{\texttt{Portfolio}} \cdots \xrightarrow{\text{merge } m} \boldsymbol{\mathcal{S}}_m^{\texttt{Portfolio}} \xrightarrow{\text{merge } m+1} \boldsymbol{\mathcal{S}}_{m+1}^{\texttt{Portfolio}}$.

By the inductive hypothesis, we know that $C(\boldsymbol{\mathcal{S}}_m^{\texttt{Portfolio}}) \geq C(\boldsymbol{\mathcal{S}}^{\texttt{Singletons}})$. Suppose, in the last merge, assays $\mathcal{S}$ and $\mathcal{S}'$ of $\boldsymbol{\mathcal{S}}_m^{\texttt{Portfolio}}$ are combined into assay $\mathcal{S} \cup \mathcal{S}'$. First consider the case where $\mathcal{S} = \{k\}$ and $\mathcal{S}' = \{\ell\}$ are singletons. As shown above (which is included in the base case), this merge cannot yield a lower cost portfolio. That is,

$$C\big((\boldsymbol{\mathcal{S}}_m^{\texttt{Portfolio}} \setminus \{\{k\}, \{\ell\}\}) \cup \{\{k, \ell\}\}\big) \ \geq\ C(\boldsymbol{\mathcal{S}}_m^{\texttt{Portfolio}}) \ \geq\ C(\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}),$$

where the last inequality follows from the inductive hypothesis.

We now extend this argument to the case where the merged assays are not necessarily singletons. Given the separability of the cost function, the merge operation affects only the cost contributions of the merged assays. Thus, the cost difference $C(\boldsymbol{\mathcal{S}}_{m+1}^{\texttt{Portfolio}}) - C(\boldsymbol{\mathcal{S}}_m^{\texttt{Portfolio}})$ is:

$$\min_{t_{\mathcal{S}\cup\mathcal{S}'}} \left\{ \sum_{r \in \mathcal{S}\cup\mathcal{S}'} \left[\frac{1}{t_{\mathcal{S}\cup\mathcal{S}'}} + 1 - (\gamma_r)^{t_{\mathcal{S}\cup\mathcal{S}'}} \right] \right\} - \min_{t_{\mathcal{S}}} \left\{ \sum_{i \in \mathcal{S}} \left[\frac{1}{t_{\mathcal{S}}} + 1 - (\gamma_i)^{t_{\mathcal{S}}} \right] \right\} - \min_{t_{\mathcal{S}'}} \left\{ \sum_{j \in \mathcal{S}'} \left[\frac{1}{t_{\mathcal{S}'}} + 1 - (\gamma_j)^{t_{\mathcal{S}'}} \right] \right\}.$$

Again, since $\mathcal{S} \cap \mathcal{S}' = \emptyset$, writing the first minimization problem as,

$$\min_{t_{\mathcal{S}}, t_{\mathcal{S}'}} \left\{ \sum_{i \in \mathcal{S}} \left[\frac{1}{t_{\mathcal{S}}} + 1 - (\gamma_i)^{t_{\mathcal{S}}} \right] + \sum_{j \in \mathcal{S}'} \left[\frac{1}{t_{\mathcal{S}'}} + 1 - (\gamma_j)^{t_{\mathcal{S}'}} \right] :\ t_{\mathcal{S}} = t_{\mathcal{S}'} \right\},$$

ensures the cost difference is non-negative. Thus, $C(\boldsymbol{\mathcal{S}}_{m+1}^{\texttt{Portfolio}}) \geq C(\boldsymbol{\mathcal{S}}_m^{\texttt{Portfolio}}) \geq C(\boldsymbol{\mathcal{S}}^{\texttt{Singletons}})$. $\square$

Proof of Proposition 3 *Part 1.* By Theorem 3, if $c_v = 0$ and $\pi(\mathcal{N}) \leq 1 - e^{-0.5}$, then $\boldsymbol{\mathcal{S}}^{n\text{-plex}}$ is optimal. We show that the optimal cost function, provided below, is continuous in c_v:

$$C^*(c_v) := \min_{\boldsymbol{\mathcal{S}}^{\text{Portfolio}} \in \mathcal{P}(\mathcal{N})} \min_{\boldsymbol{t} \in \mathcal{T}(\boldsymbol{\mathcal{S}}^{\text{Portfolio}})} C(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{t}; c_v, c_f), \text{ where} \tag{40}$$

$$C(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{t}; c_v, c_f) = c_f \cdot \Big(\big(\sum_{k \in [q]} \frac{1}{t_k} \big) + 1 - p_0 \cdot \prod_{k \in [q]} \big(p_0(\mathcal{S}_k) \big)^{t_k - 1} \Big) + c_v \cdot \Big(\sum_{i \in \mathcal{N}} \Big[\frac{1}{w_i(\boldsymbol{t})} + 1 - (1 - \pi_i)^{w_i(\boldsymbol{t})} \Big] \Big),$$

and $\mathcal{P}(\mathcal{N})$, the set of all possible partitions of $\mathcal{N}$, is a finite discrete set, and $\mathcal{T}(\boldsymbol{\mathcal{S}}^{\text{Portfolio}})$, the domain of pool sizes, is a compact set (due to the existence of a finite pool size limit $t_{\max}$).

The proof proceeds in two steps. (i) We fix a portfolio $\boldsymbol{\mathcal{S}}^{\text{Portfolio}}$ and focus on the inner minimization in (40). $C(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{t}; c_v, c_f)$ is continuous in c_v, because the first part is fixed in c_v, and the second part is linear in c_v (with coefficients depending continuously on $\boldsymbol{t}$). Then, since the domain of $\boldsymbol{t}$ is compact, Berge's Theorem of the Maximum (Berge 1963), applied to minimization, ensures that the optimal value function for the inner minimization is continuous in c_v. (ii) Since this result holds for any given portfolio and $\mathcal{P}(\mathcal{N})$ is finite, we obtain the continuity for the optimal value of the outer problem, i.e., for $C^*(c_v)$, because the minimum of finitely many continuous functions is also continuous. Hence, there exists a neighborhood of $c_v = 0$ such that $\boldsymbol{\mathcal{S}}^{n\text{-plex}}$ remains optimal.

Part 2. By Theorem 3, if $c_f = 0$ then $\boldsymbol{\mathcal{S}}^{\text{Singletons}}$ is optimal. The proof is similar to the first part, except that c_v is fixed and the optimal cost function is analyzed wrt c_f, and is omitted. □

B.6. Proofs for §3.3.2

B.6.1. Supporting Results

Lemma B.8 (Cost bounds for given marginals). *Consider any marginal prevalence vector $\boldsymbol{\pi}$ such that $\sum_{i \in \mathcal{N}} \pi_i \leq 1$. The cost for portfolio $\boldsymbol{\mathcal{S}}^{Portfolio}$ and pool size vector $\boldsymbol{t}$ for any joint vector $\boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$ can be bounded as follows:*

$$C(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}, \boldsymbol{p}^{nested}(\boldsymbol{\pi})) \leq C(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}, \boldsymbol{p}) \leq C(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}, \boldsymbol{p}^{nc}(\boldsymbol{\pi})).$$

Proof Consider any q-partition $\boldsymbol{\mathcal{S}}^{\text{Portfolio}}$. The cost of any individually tested assay does not depend on $\boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$; further, individually tested assays do not affect the cost for the pooled assays. Therefore, we consider, wlog, that $\boldsymbol{t} \geq \mathbf{2}$. For any $\boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$, where $\sum_{i \in \mathcal{N}} \pi_i \leq 1$:

$$\begin{aligned} C(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{t}, \boldsymbol{p}) = & \underbrace{\sum_{k \in [q]} \frac{c_f + s_k c_v}{t_k}}_{C_{pool}(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{t})} + \underbrace{c_f \cdot \Big(1 - (1 - \pi(\mathcal{N})) \cdot \prod_{k \in [q]} \big(1 - \pi(\mathcal{S}_k)\big)^{t_k - 1} \Big)}_{C_{fixed}(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{t}, \boldsymbol{p})} \\ & + \underbrace{c_v \cdot \Big(n - \sum_{k \in [q]} \sum_{i \in \mathcal{S}_k} (1 - \pi_i)^{t_k} \Big)}_{C_{var}(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{t}, \boldsymbol{\pi})}, \end{aligned}$$

where only C_{fixed} is a function of $\boldsymbol{p}$. Thus, maximizing (minimizing) $C(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{t}, \boldsymbol{p})$ wrt $\boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$ is equivalent to maximizing (minimizing) C_{fixed}, which requires minimizing (maximizing) $(1 - \pi(\mathcal{N})) \cdot \prod_{k \in [q]} (1 - \pi(\mathcal{S}_k))^{t_k - 1}$. This is equivalent to maximizing (minimizing) each of $\pi(\mathcal{N})$ and $\pi(\mathcal{S}_k)$, $\forall k \in [q]$, under the constraint, $\max_{i \in \mathcal{S}} \{\pi_i\} \leq \pi(\mathcal{S}) \leq \sum_{i \in \mathcal{S}} \pi_i$, $\forall \mathcal{S} \subseteq \mathcal{N}$. The bounds hold since the joint vector $\boldsymbol{p}^{\text{nc}}(\boldsymbol{\pi})$ attains the upper bound $\pi(\mathcal{S}) = \sum_{i \in \mathcal{S}} \pi_i$, $\forall \mathcal{S} \subseteq \mathcal{N}$ (maximizing C_{fixed}), while the joint vector $\boldsymbol{p}^{\text{nested}}(\boldsymbol{\pi})$ attains the lower bound (approaching $\max_{i \in \mathcal{S}} \{\pi_i\}$ as $\epsilon \to 0$) for all sets (minimizing C_{fixed}). □

Let $C^*(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{p}) := \min_{\boldsymbol{t}} C(\boldsymbol{\mathcal{S}}^{\text{Portfolio}}, \boldsymbol{t}, \boldsymbol{p})$, i.e., the adaptive cost for $\boldsymbol{\mathcal{S}}^{\text{Portfolio}}$ at its optimal pool size vector under joint vector $\boldsymbol{p}$.

B.6.2. Main Results

Proof of Theorem 4 *Part 1.* Consider any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$, with $\boldsymbol{t}^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{p})$ denoting its optimal pool size vector at $\boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$. For simplicity, we drop the argument $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$ in the functions below. For $\boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$:

$$
\begin{aligned}
C^*(\boldsymbol{p}) = C(\boldsymbol{t}^*(\boldsymbol{p}), \boldsymbol{p}) &\geq C(\boldsymbol{t}^*(\boldsymbol{p}), \boldsymbol{p}^{\texttt{nested}}(\boldsymbol{\pi})) && \text{(by Lemma B.8)} \\
&\geq C(\boldsymbol{t}^*(\boldsymbol{p}^{\texttt{nested}}(\boldsymbol{\pi})), \boldsymbol{p}^{\texttt{nested}}(\boldsymbol{\pi})) && \text{(by optimality of } \boldsymbol{t}^*(\boldsymbol{p}^{\texttt{nested}}(\boldsymbol{\pi})) \text{ at } \boldsymbol{p}^{\texttt{nested}}(\boldsymbol{\pi})) \\
&= C^*(\boldsymbol{p}^{\texttt{nested}}(\boldsymbol{\pi})). \\
C^*(\boldsymbol{p}^{\texttt{nc}}(\boldsymbol{\pi})) = C(\boldsymbol{t}^*(\boldsymbol{p}^{\texttt{nc}}(\boldsymbol{\pi})), \boldsymbol{p}^{\texttt{nc}}(\boldsymbol{\pi})) &\geq C(\boldsymbol{t}^*(\boldsymbol{p}^{\texttt{nc}}(\boldsymbol{\pi})), \boldsymbol{p}) && \text{(by Lemma B.8)} \\
&\geq C(\boldsymbol{t}^*(\boldsymbol{p}), \boldsymbol{p}) && \text{(by optimality of } \boldsymbol{t}^*(\boldsymbol{p}) \text{ at } \boldsymbol{p}) \\
&= C^*(\boldsymbol{p}).
\end{aligned}
$$

Thus, $C^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{p}^{\texttt{nested}}(\boldsymbol{\pi})) \leq C^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{p}) \leq C^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{p}^{\texttt{nc}}(\boldsymbol{\pi})),\ \forall \boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$.

Part 2. From Lemma B.8, we know that for any fixed design $(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t})$:

$$C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}, \boldsymbol{p}) \leq C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}, \boldsymbol{p}^{\texttt{nc}}(\boldsymbol{\pi})),\ \forall \boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi}).$$

Since this inequality holds for all $\boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})$, the inner maximization in *Adaptive-R* reduces to:

$$\max_{\boldsymbol{p} \in \mathcal{P}(\boldsymbol{\pi})} C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}, \boldsymbol{p}) = C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}, \boldsymbol{p}^{\texttt{nc}}(\boldsymbol{\pi})).$$

Substituting this into the outer minimization in *Adaptive-R* yields the equivalence. □

A result similar to the second part of Theorem 4 holds when uncertainty is on the cost parameters rather than the joint prevalence vector. Specifically, suppose c_f and c_v vary independently over compact uncertainty sets $\mathcal{C}_f$ and $\mathcal{C}_v$, respectively. Since the cost function $C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}, c_f, c_v)$ is increasing in both c_f and c_v, the robust adaptive testing design problem under cost parameter uncertainty reduces to a deterministic counterpart evaluated at the worst-case cost parameters, that is, $\min_{\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}} \left(\max_{c_f \in \mathcal{C}_f, c_v \in \mathcal{C}_v} C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}, c_f, c_v) \right) \equiv \min_{\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}} C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}, c_f^{\max}, c_v^{\max})$, where $c_f^{\max} := \max_{c_f \in \mathcal{C}_f} \{c_f\}$ and $c_v^{\max} := \max_{c_v \in \mathcal{C}_v} \{c_v\}$.

B.7. Proofs for §3.3.3

B.7.1. Supporting Results

Lemma B.9 (Pool size comparison with Dorfman for $c_v = 0$). *Let $c_v = 0$ and $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$. For any q-partition $\boldsymbol{\mathcal{S}}^{Portfolio}$ and $k \in [q]$, if $\beta_k(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k}) > -1/e$, then:*

$$\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k}) \geq t^{Dorf}(\mathcal{S}_k) \quad \textit{and} \quad \bar{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}, \boldsymbol{t}_{-k}) \leq \bar{t}^{Dorf}(\mathcal{S}_k).$$

Proof To prove this result, we use the characterization of the local minimum and local maximum pool sizes from Lemma B.6 when $\beta_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) > -1/e$. We drop the superscript $(c_v = 0)$ as well as the arguments in parentheses in places, and also let, for $k \in [q]$,

$$f_k(\boldsymbol{t}_{-k}) := \frac{\gamma(\mathcal{S}_k)}{p_0 \cdot \prod_{\ell \in [q] \setminus \{k\}} (\gamma(\mathcal{S}_\ell))^{t_\ell - 1}} \cdot \ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right) > 0$$

so that in Lemma B.6 we have:

$$\beta_k = -1/2 \cdot (f_k(\boldsymbol{t}_{-k}))^{1/2} < 0, \text{ and } \underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) = \frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot W_0(\beta_k).$$

Part 1. For any $m \in [q] \setminus \{k\}$, we have, by the chain rule,

$$\frac{\partial \underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})}{\partial t_m} = \frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot \frac{\partial W_0(\beta_k)}{\partial \beta_k} \cdot \frac{\partial \beta_k}{\partial t_m}, \text{ and}$$

$$\frac{\partial W_0(\beta_k)}{\partial \beta_k} = \frac{W_0(\beta_k)}{\beta_k \cdot [1 + W_0(\beta_k)]} > 0, \quad \text{by Corless et al. (1996)},$$

which follows because $-1 < W_0(\cdot) < 0$ (Property B.2) and $\beta_k < 0$. Observe that:

$$\frac{\partial \beta_k}{\partial t_m} = -\frac{1}{4} \cdot (f_k)^{-1/2} \cdot \frac{\gamma(\mathcal{S}_k)}{p_0} \cdot \ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right) \cdot \frac{1}{\prod_{\ell \in [q] \setminus \{k,m\}} (\gamma(\mathcal{S}_\ell))^{t_\ell - 1}} \cdot \frac{-(\gamma(\mathcal{S}_m))^{1-t_m}}{\ln(\gamma(\mathcal{S}_m))} < 0.$$

Thus, $\frac{\partial \underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})}{\partial t_m} > 0$, i.e., $\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})$ is increasing in any $t_m, m \neq k$. Then, to prove $\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) \geq t^{\texttt{Dorf}}(\mathcal{S}_k)$, it suffices to prove $\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k} = \mathbf{1}) \geq t^{\texttt{Dorf}}(\mathcal{S}_k)$.

Recall that $\gamma(\mathcal{S}_k) = 1 - \pi(\mathcal{S}_k) = p_0(\mathcal{S}_k)$. For $\boldsymbol{t}_{-k} = \mathbf{1}$, we have that:

$$\begin{aligned}
&\frac{\gamma(\mathcal{S}_k)}{p_0 \cdot \prod_{\ell \in [q] \setminus \{k\}} (\gamma(\mathcal{S}_\ell))^{1-1}} = \frac{\gamma(\mathcal{S}_k)}{p_0} \geq 1 \text{ (since } \pi(\mathcal{S}_k) \leq \pi(\mathcal{N}) \leq 1 - p_0) \\
&\Rightarrow (f_k(\boldsymbol{t}_{-k} = \mathbf{1}))^{1/2} = \left(\frac{\gamma(\mathcal{S}_k)}{p_0} \cdot \ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2} \geq \left(\ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2} \\
&\Rightarrow \beta_k = -\frac{1}{2} \cdot \left(\frac{\gamma(\mathcal{S}_k)}{p_0} \cdot \ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2} \leq -\frac{1}{2} \cdot \left(\ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2} \\
&\Rightarrow W_0(\beta_k) \leq W_0\left(-\frac{1}{2} \cdot \left(\ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2}\right) \text{ (by Property B.2)} \\
&\Rightarrow \overbrace{\frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot W_0(\beta_k)}^{= \underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}=\mathbf{1})} \geq \overbrace{\frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot W_0\left(-\frac{1}{2} \cdot \left(\ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2}\right)}^{= t^{\texttt{Dorf}}(\mathcal{S}_k) \text{ by Proposition B.1}} \\
&\Rightarrow \underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) \geq \underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k} = \mathbf{1}) \geq t^{\texttt{Dorf}}(\mathcal{S}_k), \ \forall \boldsymbol{t}_{-k}. \qquad \square
\end{aligned}$$

Part 2. Next, we show the relation between the finite local maxima. From Lemma B.6, we have $\bar{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) = \frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot W_{-1}(\beta_k)$. For any $m \in [q] \setminus \{k\}$, we have, by the chain rule,

$$\frac{\partial \bar{t}_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \bar{\boldsymbol{t}}_{-k})}{\partial t_m} = \frac{2}{\ln(\gamma(\mathcal{S}_k))} \cdot \frac{\partial W_{-1}(\beta_k)}{\partial \beta_k} \cdot \frac{\partial \beta_k}{\partial t_m},$$

where, by Corless et al. (1996), because $W_{-1}(\cdot) < -1$ (Property B.2) and $\beta_k < 0$, we have

$$\frac{\partial W_{-1}(\beta_k)}{\partial \beta_k} = \frac{W_{-1}(\beta_k)}{\beta_k \cdot [1 + W_{-1}(\beta_k)]} < 0.$$

From Part 1 we know that $\frac{\partial \beta_k}{\partial t_m} < 0$. Thus, $\frac{\partial \bar{t}_k}{\partial t_m} < 0$, that is, $\bar{t}_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})$ is decreasing in any $t_m, m \neq k$. To show that $\bar{t}_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) \geq t^{\texttt{Dorf}}(\mathcal{S}_k)$, it is sufficient to show that $\bar{t}_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k} = \boldsymbol{t}_{\texttt{max}}) \geq t^{\texttt{Dorf}}(\mathcal{S}_k)$, where $t_{\texttt{max}}$ is sufficiently large. We proceed as in Part 1:

$$\begin{aligned}
&\frac{\gamma(\mathcal{S}_k)}{p_0 \cdot \prod_{\ell \in [q] \setminus \{k\}} (\gamma(\mathcal{S}_\ell))^{t_{\texttt{max}} - 1}} > \frac{\gamma(\mathcal{S}_k)}{p_0} \geq 1 \\
&\Rightarrow (f_k(\boldsymbol{t}_{-k} = \boldsymbol{t}_{\texttt{max}}))^{1/2} = \left(\frac{\gamma(\mathcal{S}_k)}{p_0 \cdot \prod_{\ell \in [q] \setminus \{k\}} (\gamma(\mathcal{S}_\ell))^{t_{\texttt{max}} - 1}} \cdot \ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2} > \left(\ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2}
\end{aligned}$$

$$\Rightarrow \beta_k = -\frac{1}{2}\cdot\left(\frac{\gamma(\mathcal{S}_k)}{p_0\cdot\prod_{\ell\in[q]\setminus\{k\}}(\gamma(\mathcal{S}_\ell))^{t_{\max}-1}}\cdot\ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2} < -\frac{1}{2}\cdot\left(\ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2}$$

$$\Rightarrow W_{-1}(\beta_k) < W_{-1}\left(-\frac{1}{2}\cdot\left(\ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2}\right) \text{ (by Property B.2)}$$

$$\Rightarrow \underbrace{\frac{2}{\ln(\gamma(\mathcal{S}_k))}\cdot W_{-1}(\beta_k)}_{\bar{t}_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}_{-k})=\boldsymbol{t}_{\max}} < \underbrace{\frac{2}{\ln(\gamma(\mathcal{S}_k))}\cdot W_{-1}\left(-\frac{1}{2}\cdot\left(\ln\left(\frac{1}{\gamma(\mathcal{S}_k)}\right)\right)^{1/2}\right)}_{\bar{t}^{\texttt{Dorf}}(\mathcal{S}_k) \text{ by Proposition B.1}}$$

$$\Rightarrow \bar{t}_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}_{-k}) \le \bar{t}_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}_{-k}=\boldsymbol{t}_{\max}) < \bar{t}^{\texttt{Dorf}}(\mathcal{S}_k),\ \forall \boldsymbol{t}_{-k}, \text{ completing the proof.} \quad \square$$

Proposition B.3 (Portfolio-specific pool size bounds). *Consider that $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$. For any q-partition $\boldsymbol{\mathcal{S}}^{Portfolio}$, if the optimal pool size for any assay $k\in[q]$ satisfies the FOC, then it can be bounded as follows for any $\boldsymbol{t}_{-k}$:*

1. $\beta_k(\boldsymbol{\mathcal{S}}^{Portfolio},\boldsymbol{t}_{-k}) > -1/e \Rightarrow \min\left\{\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{Portfolio}), t^{Dorf}(\{i_1^k\})\right\} \le t_k^*(\boldsymbol{\mathcal{S}}^{Portfolio}) \le \bar{t}^{Dorf}(\{i_{s_k}^k\})$,
2. $\beta_k(\boldsymbol{\mathcal{S}}^{Portfolio},\boldsymbol{t}_{-k}) \le -1/e \Rightarrow t^{Dorf}(\{i_1^k\}) \le t_k^*(\boldsymbol{\mathcal{S}}^{Portfolio}) \le \bar{t}^{Dorf}(\{i_{s_k}^k\})$,

where i_1^k and $i_{s_k}^k$ resp. correspond to the highest and lowest marginal prevalence diseases in $\mathcal{S}_k$.

Proof Consider any q-partition $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$. From Theorem 1,

$$C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}) = \underbrace{c_f\cdot\left(\left(\sum_{k\in[q]}\frac{1}{t_k}\right)+1-p_0\cdot\prod_{k\in[q]}\left(\gamma(\mathcal{S}_k)\right)^{t_k-1}\right)}_{C^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})} + \underbrace{c_v\cdot\left(\sum_{k\in[q]}\sum_{i\in\mathcal{S}_k}\left[\frac{1}{t_k}+1-(\gamma_i)^{t_k}\right]\right)}_{C^{(c_f=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})},$$

where $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ is either a boundary solution (equaling $t_{\max}$) or satisfies the FOC. In the following we establish bounds for the case where it corresponds to an FOC.

First consider the case where $\beta_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}_{-k}) > -1/e$ for some $k\in[q]$, that is, the FOC pool size exists for function $C^{(c_v=0)}(t_k,\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}_{-k})$ when $c_v=0$, and the local minimum pool size is given by $\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ (Lemma B.6). The following properties hold:

- $C^{(c_v=0)}(t_k,\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}_{-k})$ is decreasing in t_k for $t_k < \underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ and $t_k > \bar{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ (Lemma B.6).
- In $C^{(c_f=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$, each term $\frac{1}{t_k}+1-(\gamma_i)^{t_k}$ is of the same form as Dorfman cost function ((4)), hence is decreasing in t_k for $t_k < t^{\texttt{Dorf}}(\{i\})$ and $t_k > \bar{t}^{\texttt{Dorf}}(\{i\})$, $\forall i\in\mathcal{S}_k$ (Proposition B.1).

Consequently, $C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t})$ is decreasing in t_k for any t_k that satisfies that,

$$t_k < \min\left\{\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}), \min_{i\in\mathcal{S}_k} t^{\texttt{Dorf}}(\{i\})\right\} \text{ and } t_k > \max\left\{\bar{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}), \max_{i\in\mathcal{S}_k}\bar{t}^{\texttt{Dorf}}(\{i\})\right\}. \tag{41}$$

Further, by Lemma B.9 and Corollary B.1, for any $\mathcal{S}_k=\{i_1^k,i_2^k,\ldots,i_{s_k}^k\}$, with diseases indexed following a nonincreasing order of marginal disease prevalences, we know that

$$t^{\texttt{Dorf}}(\mathcal{S}_k) \le t^{\texttt{Dorf}}(\{i_1^k\}) \le t^{\texttt{Dorf}}(\{i_2^k\}) \le\cdots\le t^{\texttt{Dorf}}(\{i_{s_k}^k\}),\quad \underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \ge t^{\texttt{Dorf}}(\mathcal{S}_k),$$
$$\bar{t}^{\texttt{Dorf}}(\mathcal{S}_k) \le \bar{t}^{\texttt{Dorf}}(\{i_1^k\}) \le \bar{t}^{\texttt{Dorf}}(\{i_2^k\}) \le\cdots\le \bar{t}^{\texttt{Dorf}}(\{i_{s_k}^k\}),\quad \bar{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \le \bar{t}^{\texttt{Dorf}}(\mathcal{S}_k).$$

Then, by (41), $C(t_k,\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}},\boldsymbol{t}_{-k})$ is decreasing in t_k for any

$$t_k < \min\left\{\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}), t^{\texttt{Dorf}}(\{i_1^k\})\right\} \text{ and } t_k > \bar{t}^{\texttt{Dorf}}(\{i_{s_k}^k\}.$$

Because we consider the case where $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ is an FOC, it must be true that $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \notin (\bar{t}^{\texttt{Dorf}}(\{i_{s_k}^k\},\infty)$, $\forall k\in[q]$, hence,

$$\min\left\{\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}), t^{\texttt{Dorf}}(\{i_1^k\})\right\} \le t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \le \bar{t}^{\texttt{Dorf}}(\{i_{s_k}^k\}).$$

Next consider the case where $\beta_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) \leq -1/e$ for some $k \in [q]$, that is, the FOC does not exist for function $C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})$, that is, $\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ and $\overline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ are undefined, and function $C^{(c_v=0)}(t_k, \boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})$ is decreasing, $\forall t_k > 0$ (Lemma B.6). Then, following a similar argument to the first case, the bounds reduce to:

$$t^{\texttt{Dorf}}(\{i_1^k\}) \leq t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \leq \overline{t}^{\texttt{Dorf}}(\{i_{s_k}^k\}), \text{ completing the proof.} \quad \square$$

B.7.2. Main Results

Proof of Theorem 5 Because $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$, by Theorem 2, for any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$, $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \neq 1$, $\forall k \in [q]$, that is, pooling is optimal for all assays. Further, because $\pi(\mathcal{N}) < \underline{p}_{\min}(\boldsymbol{\pi})$ and $\underline{p}^{\texttt{Dorf}} \leq \underline{p}_{\min}(\boldsymbol{\pi})$ (Theorem 2, Proposition 1), the Dorfman pool size for any assay satisfies the FOC, and is characterized in Proposition B.1.

Consider any assay $k \in [q]$. For the lower bound on the optimal pool size for assay k, first consider the case where $\beta_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) > -1/e$. We can write the following inequalities:

$$\begin{aligned}
&\min\left\{\underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}), t^{\texttt{Dorf}}(\{i_1^k\})\right\} \leq t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) && \text{(by Proposition B.3)}\\
&t^{\texttt{Dorf}}(\mathcal{N}) \leq t^{\texttt{Dorf}}(i_1^k) && \text{(by Corollary B.1)}\\
&t^{\texttt{Dorf}}(\mathcal{N}) \leq t^{\texttt{Dorf}}(\mathcal{S}_k) \leq \underline{t}_k^{(c_v=0)}(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) && \text{(by Corollary B.1 and Lemma B.9)},
\end{aligned}$$

establishing that $t^{\texttt{Dorf}}(\mathcal{N}) \leq t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$. For the case where $\beta_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k}) \leq -1/e$, $\underline{t}_k^{(c_v=0)}$ does not exist (Lemma B.6), but because $\pi(\mathcal{N}) < \underline{p}^{\texttt{Dorf}}$, we still have that, $t^{\texttt{Dorf}}(\{i_1^k\}) \leq t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}})$ (Proposition B.3), and the result follows similarly.

For the upper bound on the optimal pool size for assay k, for all values of $\beta_k(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}_{-k})$:

$$\begin{aligned}
&t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \leq \overline{t}^{\texttt{Dorf}}(\{i_{s_k}^k\}) \quad \text{(by Proposition B.3)}\\
&\overline{t}^{\texttt{Dorf}}(\{i_{s_k}^k\}) \leq \overline{t}^{\texttt{Dorf}}(\{n\}) \quad \text{(by Property B.2 since } \pi_n \leq \pi_{i_{s_k}^k}\text{)},
\end{aligned}$$

establishing that $t_k^*(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}) \leq \overline{t}^{\texttt{Dorf}}(\{n\})$.

Then, incorporating the finite pool size limit ($t_{\texttt{max}} < \infty$) and the integrality of the pool sizes leads to the lower and upper bounds provided in the theorem. Finally, the result that $t^{\texttt{LB}} \geq 2$ follows directly from the result that $t^{\texttt{Dorf}}(\mathcal{S}) \geq 2.57$, $\forall \mathcal{S} \subseteq \mathcal{N}$ (Proposition B.1). $\square$

Proof of Theorem 6 The bound $C^{\texttt{LB}}$ is obtained by separately bounding the fixed and variable cost terms of the cost function in (5):

$$C(\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}, \boldsymbol{t}) = \overbrace{c_f \cdot \left(\Big(\sum_{k\in[q]} \frac{1}{t_k}\Big) + 1 - p_0 \cdot \prod_{k\in[q]} \big(1 - \pi(\mathcal{S}_k)\big)^{t_k - 1}\right)}^{\text{Subproblem } (c_v=0) \text{ objective}} + \overbrace{c_v \cdot \left(\sum_{k\in[q]} \sum_{i\in\mathcal{S}_k} \left[\frac{1}{t_k} + 1 - (1-\pi_i)^{t_k}\right]\right)}^{\text{Subproblem } (c_f=0) \text{ objective}}.$$

Subproblem ($c_v = 0$) and ($c_f = 0$) objectives correspond to the special cases of $c_v = 0$ and $c_f = 0$, resp., for which the optimal solutions are given in Theorem 3. In particular, $\boldsymbol{\mathcal{S}}^{n\texttt{-plex}}$ with pool size $t^{\texttt{Dorf}}(\mathcal{N})$ is optimal when $c_v = 0$. Hence, for any portfolio $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$ and pool size vector $\boldsymbol{t}$,

$$c_f \cdot \left(\Big(\sum_{k\in[q]} \frac{1}{t_k}\Big) + 1 - p_0 \cdot \prod_{k\in[q]} \big(p_0(\mathcal{S}_k)\big)^{t_k - 1}\right) \geq c_f\left(\frac{1}{t^{\texttt{Dorf}}(\mathcal{N})} + 1 - p_0^{t^{\texttt{Dorf}}(\mathcal{N})}\right).$$

Similarly, from Theorem 3, $\boldsymbol{\mathcal{S}}^{\texttt{Singletons}}$ with pool sizes $t^{\texttt{Dorf}}(\{k\}), \forall k \in [n]$, is optimal when $c_f = 0$. Hence, for any portfolio $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$ and pool size vector $\boldsymbol{t}$,

$$c_v\left(\sum_{k\in[q]} \sum_{i\in\mathcal{S}_k} \Big[\frac{1}{t_k} + 1 - (1-\pi_i)^{t_k}\Big]\right) \geq c_v\left(\sum_{i\in\mathcal{N}} \Big[\frac{1}{t^{\texttt{Dorf}}(\{i\})} + 1 - (1-\pi_i)^{t^{\texttt{Dorf}}(\{i\})}\Big]\right).$$

Because these inequalities are satisfied for any $\boldsymbol{\mathcal{S}}^{\texttt{Portfolio}}$ and $\boldsymbol{t}$, they remain valid for the optimal portfolio and its optimal pool size vector. Combining the inequalities yields the lower bound. $\square$

C. Bucket-based Formulation for Independent and Nested Structures

Independent structure requires: (1) Removing variables $\tilde{\boldsymbol{\pi}}$, $\log\tilde{\boldsymbol{\pi}}$, $\log\tilde{\boldsymbol{e}}$ and associated constraints (8f), (8l), and (8o). (2) Replacing (8m) with $\log p = \sum_{i\in\mathcal{N}} \log e_i$. **Nested structure** requires: (1) Defining variables $y_b \in \{0,1\}^n$ and $\pi_b^{\max}$, $\forall b\in\mathcal{N}$. (2) Adding the following constraints:

$$
\begin{aligned}
& y_b^i \le x_b^i && \forall i\in\mathcal{N},\ \forall b\in\mathcal{N}\\
& y_b^i \le 1 - x_b^j && \forall i\in\{2,\dots,n\},\ \forall j\in\{1,\dots,i-1\},\ \forall b\in\mathcal{N}\\
& \sum_{i\in\mathcal{N}} y_b^i = z_b && \forall b\in\mathcal{N}\\
& \pi_b^{\max} = \sum_{i\in\mathcal{N}} \pi_i\cdot y_b^i && \forall b\in\mathcal{N}
\end{aligned}
$$

(3) Replacing (8f) with $\tilde{\pi}_b = 1-\epsilon\cdot\sum_{i\in\mathcal{N}} x_b^i - \pi_b^{\max} + \epsilon z_b,\quad \forall b\in\mathcal{N}$.

D. Supplementary Numerical Analysis and Case Study Details

D.1. Supplementary Numerical Analysis

Example 4 (**Nonmonotonicity of adaptive pool size in assay prevalence**). Consider an instance with $n=4$, $c_f=20, c_v=5$, and marginal prevalences $\pi_1(\theta)=0.067851\,\theta$ and $\pi_2(\theta)=0.053303\,\theta$ (parameterized by θ), $\pi_3=0.042999$, and $\pi_4=0.039108$ under the no-coinfection structure. Consider the portfolio $\mathcal{S}_1=\{1,2\}$ and $\mathcal{S}_2=\{3,4\}$, with assay prevalences $\pi(\mathcal{S}_1(\theta))=\pi_1(\theta)+\pi_2(\theta)=0.121154\,\theta$ and $\pi(\mathcal{S}_2)=\pi_3+\pi_4\approx 0.082107$. For $\theta\in\{0.6,\,1.0,\,1.6,\,2.2\}$, Table B.1 reports the optimal adaptive pool sizes $t_1^*(\mathcal{S}_1,\theta), t_2^*(\mathcal{S}_2,\theta)$ (confirmed to be unique) and optimal cost $C^*(\mathcal{S}_1,\mathcal{S}_2,\theta)$, showing that $t_1^*(\mathcal{S}_1,\theta)$ decreases as $\pi(\mathcal{S}_1(\theta))$ increases from 0.072692 to 0.193846, but then increases (from 4.340 to 4.365, bolded) as $\pi(\mathcal{S}_1(\theta))$ rises further to 0.266539. Thus, the optimal adaptive pool size need not be monotone in assay prevalence.

Table B.1 Optimal adaptive pool sizes and cost for portfolio $\mathcal{S}_1=\{1,2\},\mathcal{S}_2=\{3,4\}$ (no-coinfection structure).

θ	$\pi(\mathcal{S}_1(\theta))$	$t_1^*(\mathcal{S}_1,\theta)$	$t_2^*(\mathcal{S}_2,\theta)$	$C^*(\mathcal{S}_1,\mathcal{S}_2,\theta)$
0.6	0.072692	5.740	5.400	26.611609
1.0	0.121154	4.820	5.845	29.701722
1.6	0.193846	**4.340**	6.580	33.078117
2.2	0.266539	**4.365**	7.520	35.569923

Monte Carlo Simulation (§5.3) In our simulations, we independently vary test specificity (0.95–1.00) and sensitivity (0.95–1.00). For each simulation run, we generate 1 million subjects, randomly assigning each subject to a disease category based on the given joint prevalence vector. To determine the cost of a specific testing design (comprising a portfolio and pool sizes), we construct random pools for each assay in the portfolio by randomly assigning the subjects according to the assay's pool size, independently of all other pools. We then simulate the test outcome for each pool based on the true disease categories of the subjects within the pool and the provided test sensitivity and specificity (using uniformly distributed realizations), and compute the testing cost.

D.2. Case Study Details

A Bayesian network (BN) is a directed acyclic graph (Pearl 2014) suitable for modeling the effect of seasonality on respiratory disease prevalences. We use the simple BN shown in Fig. B.1, which captures seasonal dependencies through the month variable. To estimate the joint prevalence vector, we employ a forward sampling approach (Koller and Friedman 2009) to generate 10 million samples, each representing the disease status of a random subject, as follows: (1) We sample a month $\tau \in \{1, 2, \cdots, 12\}$. (2) Given the month, we determine the presence/absence of each disease in the subject via independently sampled Bernoulli distributions, each with success probability $\pi_{i,\tau}$ or $\hat{\pi}_{i,\tau}$ (for $i \in \mathcal{N}$) in the accurate- versus inaccurate-mean scenarios. Using the 10 million subjects, we compute the frequency of each disease category to derive the annual joint prevalence vector.

Table B.2 Yearly marginal prevalences for 2018–2021 (disease indexing based on 2021 prevalences).

Disease	**Index**	**2018**	**2019**	**2020**	**2021**
Respiratory syncytial virus (RSV)	1	6.15%	6.67%	2.05%	6.16%
COVID-19	2	N/A	N/A	3.59%	6.03%
Parainfluenza virus 3	3	2.75%	2.73%	0.12%	3.38%
Respiratory adenovirus	4	3.08%	3.66%	1.86%	2.84%
Human metapneumovirus	5	3.15%	2.94%	1.50%	1.46%
CoVOC43	6	1.34%	1.35%	0.23%	1.34%
Parainfluenza virus 2	7	0.93%	0.21%	0.05%	0.77%
CoVNL63	8	0.70%	0.70%	0.53%	0.71%
Mycoplasma pneumoniae	9	19.10%	29.46%	9.12%	0.53%
Influenza A[1]	10	0.51%	0.49%	0.50%	0.50%
Parainfluenza virus 4	11	0.62%	0.54%	0.22%	0.48%
CoV229E	12	0.27%	0.27%	0.07%	0.28%
Bordetella parapertussis	13	0.21%	0.21%	0.21%	0.21%
Bordetella pertussis	14	0.04%	0.04%	0.04%	0.04%
Chlamydophila pneumoniae	15	0.03%	0.03%	0.03%	0.04%
Parainfluenza virus 1	16	0.20%	1.51%	0.16%	0.03%
CoVHKU1	17	0.03%	0.03%	0.72%	0.03%
Influenza B	18	9.84%	8.83%	4.87%	0.01%

[1] Influenza A prevalence represents the sum of the prevalences of its subtypes H1 and H2.

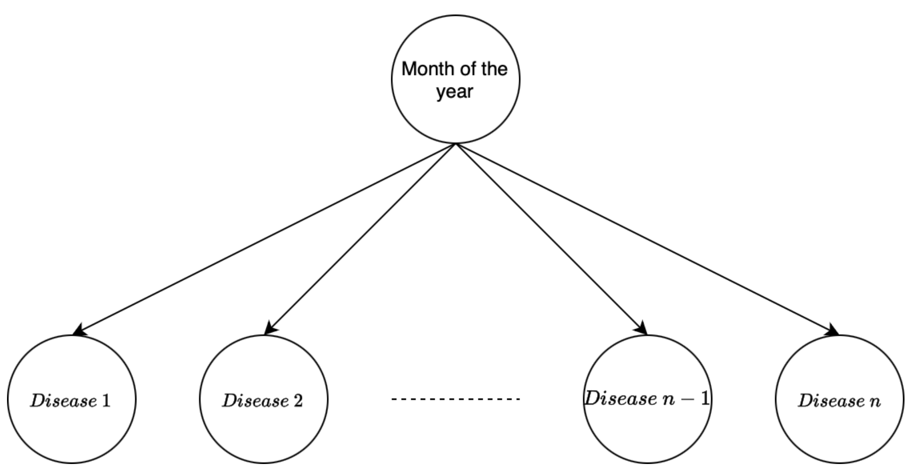
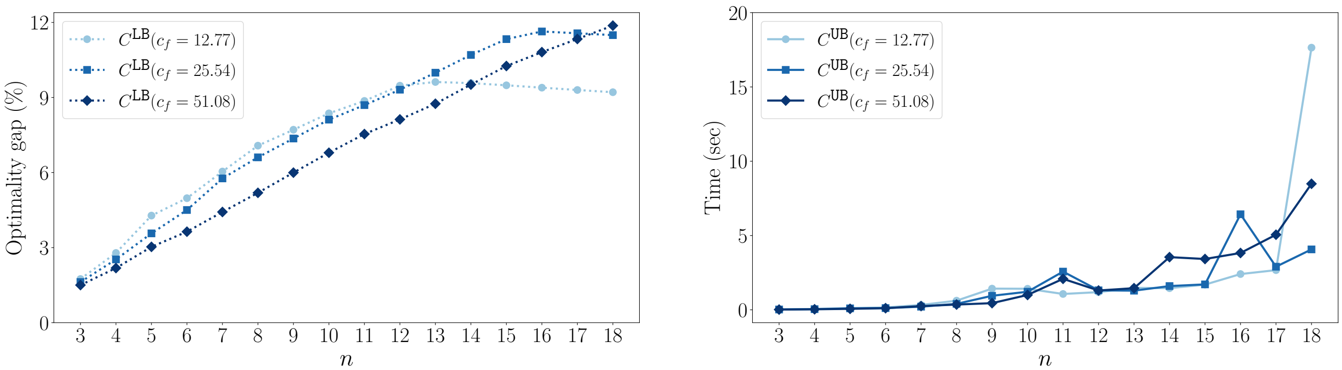


Figure B.1 Bayesian network for joint prevalence vector estimation.

D.3. Additional Case Study Results

We conduct a stress test to examine the sensitivity of annual testing costs to the assumed structure of the actual prevalence vector (i.e., independent infections within each month) used for design evaluation.



(a) Optimality gaps (%) for C^{LB} (b) Computational times (seconds) for C^{UB}

Figure B.2 (a) Optimality gaps for the lower bound C^{LB}, and (b) computational times for the upper bound C^{UB}.

The decision-maker continues to optimize based on the joint prevalences generated by the BN, but the resulting annual designs are re-evaluated under two extreme correlation structures: no coinfections and nested (Definition 1), with monthly marginal prevalences, $\boldsymbol{\pi}_\tau$, held fixed at the values obtained from the CDC dataset. The results, reported in Table B.3, for the accurate-mean setting for 2021 designs, indicate that adaptive designs continue to perform well across all joint prevalence structures considered. Moreover, even under extreme shifts in the prevalence structure, adaptive designs yield more robust costs than the benchmarks.

Table B.3 Accurate-mean setting: Annual testing costs for 2021 designs under different actual prevalence structures.

	Adaptive Designs		**Benchmarks**		
Prevalence Structure	*Adaptive-D*	*Adaptive-R*	*Dorfman*	*Dorfman-n-plex*	*Individual-n-plex*
Independent	41.33	41.16	65.84	90.02	105.82
No-coinfection	41.90	41.68	67.49	94.16	105.82
Nested	34.05	34.07	48.91	61.82	105.82

Table B.4 Accurate- and inaccurate-mean settings: 2019 designs under assay size limit $s_{\max} = 12$.

Design (2019 accurate-mean)	**Portfolio**	**Pool Sizes**	**Cost**
Adaptive-D	[1, 3–7, 9, 18], [8, 10–17]	8, 16	49.85
Adaptive-R	[1, 3–7, 9, 18], [8, 10–17]	8, 16	49.85
Design (2019 inaccurate-mean)	**Portfolio**	**Pool Sizes**	**Cost**
Adaptive-D	[1, 3–7, 9, 18], [8, 10–17]	7, 16	50.38
Adaptive-R	[1, 3–6, 9, 18], [7, 8, 10–17]	7, 16	50.10

Table B.5 Accurate-mean setting: 2021 and 2019 designs.
(All designs are ordered partitions based on monthly prevalences).

Design	Portfolio	Pool Sizes
	— 2021 Designs —	
Adaptive-D	[1–9], [10–18]	6, 16
Adaptive-R	[1–8], [9–18]	6, 16
Dorfman	[1–6], [7–12], [13–18]	3, 6, 16
Adaptive-PI (Jan)	[2, 4, 10], [1, 3, 5–9, 11–18]	4, 16
Adaptive-PI (Feb)	[2, 4, 8, 10], [1, 3, 5–7, 9, 11–18]	5, 16
Adaptive-PI (Mar)	[1–4, 6, 8, 10], [5, 7, 9, 11–18]	6, 16
Adaptive-PI (Apr)	[1–4, 6, 8, 10], [5, 7, 9, 11–18]	5, 16
Adaptive-PI (May)	[1–4, 6–8, 10], [5, 9, 11–18]	5, 16
Adaptive-PI (Jun)	[1–4, 6–8], [5, 9–18]	5, 16
Adaptive-PI (Jul–Aug)	[1–4, 6, 7], [5, 8–18]	5, 16
Adaptive-PI (Sep)	[1–5, 7], [6, 8–18]	5, 16
Adaptive-PI (Oct)	[1–5, 7, 11], [6, 8–10, 12–18]	5, 16
Adaptive-PI (Nov)	[1–7, 9, 11], [8, 10, 12–18]	5, 16
Adaptive-PI (Dec)	[1–7, 9, 11], [8, 10, 12–18]	6, 16
Dorfman-PI (Jan)	[2, 4], [8, 10, 13], [1, 5–7, 11], [3, 9, 12, 14–18]	3, 11, 16, 16
Dorfman-PI (Feb)	[2, 4, 8, 10], [1, 3, 6, 7, 12, 13], [5, 9, 11, 14–18]	4, 12, 16
Dorfman-PI (Mar)	[2, 4, 8, 10], [1, 3, 6, 7, 12, 13], [5, 9, 11, 14–18]	4, 8, 16
Dorfman-PI (Apr)	[1–4, 6, 8], [7, 10, 12, 13], [5, 9, 11, 14–18]	3, 9, 16
Dorfman-PI (May)	[1–4, 6, 8], [5, 7, 10, 12, 13], [9, 11, 14–18]	3, 8, 16
Dorfman-PI (Jun–Jul)	[1–4, 6], [5, 7, 8, 10, 12, 13], [9, 11, 14–18]	3, 7, 16
Dorfman-PI (Aug)	[1–4], [5–7, 10], [8, 11–13], [9, 14–18]	3, 6, 10, 16
Dorfman-PI (Sep)	[1–4, 7], [5, 6, 10–13], [8, 9, 14–18]	3, 7, 16
Dorfman-PI (Oct)	[1–5, 7], [6, 9–13], [8, 14–18]	3, 7, 16
Dorfman-PI (Nov)	[1–5, 7, 9, 11], [6, 10, 12, 13], [8, 14–18]	3, 8, 16
Dorfman-PI (Dec)	[1–5, 9], [6, 7, 10–12], [8, 13–18]	3, 5, 14
	— 2019 Designs —	
Adaptive-D	[1, 3–18]	11
Adaptive-R	[1, 3–18]	11
Dorfman	[1, 3–6, 9, 16, 18], [8, 10–13], [7, 14, 15, 17]	1, 7, 16
Adaptive-PI (Jan)	[1, 3–18]	15
Adaptive-PI (Feb)	[1, 3–18]	16
Adaptive-PI (Mar)	[1, 3–18]	14
Adaptive-PI (Apr)	[1, 3–18]	11
Adaptive-PI (May)	[1, 3–6, 8, 9, 18], [7, 10–17]	6, 16
Adaptive-PI (Jun)	[1, 3–6, 9, 18], [7, 8, 10–17]	6, 16
Adaptive-PI (Jul)	[1, 3–6, 9, 16, 18], [7, 8, 10–15, 17]	6, 16
Adaptive-PI (Aug)	[1, 3–18]	11
Adaptive-PI (Sep)	[1, 4, 9, 16, 18], [3, 5–8, 10–15, 17]	5, 16
Adaptive-PI (Oct)	[1, 4, 9, 11, 16, 18], [3, 5–8, 10, 12–15, 17]	5, 16
Adaptive-PI (Nov)	[1, 3–18]	12
Adaptive-PI (Dec)	[1, 3–18]	13
Dorfman-PI (Jan)	[1, 4, 5, 9], [3, 7, 10, 11, 18], [8, 13, 16], [6, 12, 14, 15, 17]	1, 5, 15, 16
Dorfman-PI (Feb)	[3–5, 9, 18], [7, 8, 10, 11, 13], [1, 6, 12, 14–17]	1, 8, 16
Dorfman-PI (Mar)	[3–5, 8, 9, 18], [1, 6, 7, 10, 12, 13], [11, 14–17]	1, 8, 16
Dorfman-PI (Apr)	[3–6, 8, 9, 18], [1, 10, 12, 13, 16], [7, 11, 14, 15, 17]	1, 8, 16
Dorfman-PI (May)	[1, 3–6, 8, 9, 18], [10–13, 16], [7, 14, 15, 17]	1, 8, 16
Dorfman-PI (Jun–Jul)	[1, 3–6, 9, 18], [8, 10–12, 16], [7, 13–15, 17]	1, 7, 16
Dorfman-PI (Aug)	[1, 4, 9, 18], [3, 5, 6, 10, 16], [8, 11–13], [7, 14, 15, 17]	1, 5, 10, 16
Dorfman-PI (Sep)	[1, 4, 9, 16, 18], [3, 5, 6, 10–13], [7, 8, 14, 15, 17]	1, 6, 16
Dorfman-PI (Oct)	[1, 4, 9, 16, 18], [3, 5, 6, 10, 11, 13], [7, 8, 12, 14, 15, 17]	1, 6, 15
Dorfman-PI (Nov)	[1, 4, 5, 9, 11, 16, 18], [3, 6, 10, 12, 13], [7, 8, 14, 15, 17]	1, 8, 16
Dorfman-PI (Dec)	[1, 4–6, 9, 11, 16, 18], [3, 10, 12, 13], [7, 8, 14, 15, 17]	1, 9, 16