MARGINAL ASYMPTOTICS FOR THE “LARGE P, SMALL N” PARADIGM: WITH APPLICATIONS TO MICROARRAY DATA

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The “large p, small n” paradigm arises in microarray studies, where expression levels of thousands of genes are monitored for a small number of subjects. There has been an increasing demand for study of asymptotics for the various statistical models and methodologies using genomic data. In this article, we focus on one-sample and two-sample microarray experiments, where the goal is to identify significantly differentially expressed genes. We establish uniform consistency of certain estimators of marginal distribution functions, sample means and sample medians under the large p small n assumption. We also establish uniform consistency of marginal p-values based on certain asymptotic approximations which permit inference based on false discovery rate techniques. The affects of the normalization process on these results is also investigated. Simulation studies and data analyses are used to assess finite sample performance.

Key words and phrases. Brownian bridge, Brownian motion, Empirical Process, False discovery rate, Hungarian construction, Marginal asymptotics, Maximal inequalities, Microarrays.
1. Introduction. Microarrays are capable of monitoring gene expression on a large scale and are becoming a routine tool in biomedical research. Studies of associations between microarray measurements and variations of phenotypes can lead to a better treatment assignment and so there has been an increasing demand for novel statistical tools analyzing such data. For example, several recent developments in microarray analysis have involved semiparametric model methodology. Such research includes, but is not limited to, estimation of normalization effects with a semi-linear in-slide model (SLIM) in Fan, Peng and Huang (2004) (FPH hereafter), estimation and inference of gene effects in Yang et al. (2001) and Huang, Wang and Zhang (2005) (HWZ hereafter), classification of phenotypes based on Affymetrix genechip data in Ghosh and Chinnaiyan (2004), and survival analysis with right censored data and genomic covariates (Gui and Li, 2004).

Although statistical analysis with microarray data has been one of the most investigated areas, theoretical studies of asymptotic properties of different statistical methodologies remain rare (for important exceptions to this, see van der Laan and Bryan, 2001; FPH; and HWZ). The paucity of such research is partly caused by the abnormal type of asymptotics associated with microarrays: the dimension of the covariate $p$ is usually much larger than the sample size $n$, i.e., the "large p, small n" paradigm referred to in West (2003). In this article, we focus on asymptotics for the simple settings of one-sample and two-sample comparisons, where the goal is to find genes differentially expressed for different phenotype groups.

Consider, for example, a simple one-sample cDNA microarray study, where the goal is to identify genes differentially expressed from zero. Note that this data setting and the following discussions can be easily extended to incorporate two-sample microarray studies as in Yang et al. (2001). Studies using Affymetrix genechip data can be included in the same framework with only minor modifications. Denote $Y_{ij}$ and $Z_{ij}$ as the background-corrected log-ratios and log-intensities (as in HWZ), for array $i = 1, \ldots, n$ and gene $j = 1, \ldots, p$. We consider the following simplified partial linear model for cDNA microarray data:

\begin{equation}
Y_{ij} = \mu_j + h_i(Z_{ij}) + \epsilon_{ij},
\end{equation}
where $\mu_j$ are the fixed gene effects, $h_i(Z_{ij})$ are the smooth array-specific normalization effects (constrained to have mean zero within each array) and $\epsilon_{ij}$ are mean zero (within array) random errors. The constraints are for model identifiability. For simplicity of exposition, we have omitted other potentially important terms in our model, such as possible print-tip effects, and array-specific position and scale constants. We note, however, that the theory we present in this paper can extend readily to these richer models.

Models similar to (1) have been investigated by HWZ and FPH. In HWZ, asymptotic properties based on least squares estimation are established assuming fixed $p$ and $n \to \infty$. It is shown that $\mu_j$ and $h_i$ can both be consistently estimated with optimal convergence rates. In FPH, partial consistency type asymptotics are established. It is proved that when $n$ is fixed and $p \to \infty$, $h_i$ can be consistently estimated by an estimator $\hat{h}_i$, although $\mu_j$ cannot be consistently estimated. If we let $X_{ij} = \mu_i + \epsilon_{ij}$ and $\tilde{X}_{ij} = Y_{ij} - \hat{h}_i(X_{ij})$, the results of FPH can be restated as $\max_{1 \leq i \leq n} \max_{1 \leq j \leq p} |\tilde{X}_{ij} - X_{ij}| = o_P(1)$. In otherwords, the normalization process is consistent. This permits the use of the normalized array-specific gene effects $\tilde{X}_{ij}$ for inference in place of the true array-specific gene effects $X_{ij}$. However, because $n$ is fixed, the permissible inference tools at the gene level are restricted to exact methods, such as permutation tests.

The goal of our paper is to study normalization and inference when the number of arrays $n \to \infty$ slowly while the number of genes $p \gg n$. This is essentially the same asymptotic framework considered in van der Laan and Bryan (2001) who show that provided the range of expression levels is bounded, the sample means consistently estimate the mean gene effects uniformly across genes whenever $\log p = o(n)$. We extend the results of van der Laan and Bryan (2001), FPH and HWZ in three important ways. First, uniform consistency results are extended to general empirical distribution functions and sample medians. Second, a precise Brownian bridge approximation to the empirical distribution function is developed and utilized to establish uniform validity of marginal p-values based on approximations which are asymptotic in $n$. The statistical tests we consider for this purpose include both one and two sample mean and median tests as well as several other functionals of the empirical distribution function. We find that the rate requirement is either $\log p = o(n^{1/2})$ or $\log p = o(n^{1/3})$, depending on the choice of test statistic. Third,
these results are further extended to allow for the presence of normalization error. An important consequence of these results is that approximate p-values based on normalized gene expression data can be validly applied to false discovery rate (FDR) methods (see Benjamini and Hochberg, 1995) for identifying differentially expressed genes. We refer to this kind of asymptotic regime as “marginal asymptotics” (see also Kosorok and Ma, 2005) because the focus of the inference is at the marginal (gene) level, even though the results are uniformly valid over all genes. The main conclusion of our paper is that the marginal asymptotic regime is valid even if the number of genes increases almost exponentially relative to the number of arrays, i.e., log $p_n = o(n^\alpha)$ for some $\alpha > 0$. Qualitatively, this seems to be the correct order of asymptotics for microarray experiments with a moderate number, say $\sim 50$, of replications. The main tools we use to obtain these results include maximal inequalities, a specialized Hungarian construction for the empirical distribution function, and a precise bound on the modulus of continuity of Brownian motion.

The article is organized as follows. In sections 2–4, we investigate marginal asymptotics based on the true gene effects (no normalization error). Section 2 discusses one-sample inference based on the mean and the median. Section 3 extends section 2 to the two-sample setting. Section 4 considers one and two sample inference when the statistics are distribution free. Section 5 demonstrates under reasonable regularity conditions that the asymptotic results obtained in sections 2–4 are not affected by the normalization process. Simulation studies and data analyses in section 6 are used to assess the finite sample performance and to demonstrate the practical utility of the proposed asymptotic theory. A brief discussion is given in section 7. Proofs are given in section 8.

2. Marginal asymptotics for one sample studies. The results of this section are based on the true data (without normalization error). For each $n \geq 1$, let $X_{1(n)}, \ldots, X_{n(n)}$ be a sample of i.i.d. vectors of length $p_n$, where the dependence within vectors is allowed to be arbitrary. Denote the $j$th component of the $i$th vector $X_{ij(n)}$, i.e., $X_i(n) = (X_{i1(n)}, \ldots, X_{ip_n(n)})'$. Also let the marginal distribution of $X_{1j(n)}$ be denoted $F_{j(n)}$, and let $\hat{F}_{j(n)}(t) = n^{-1} \sum_{i=1}^n 1\{X_{ij(n)} \leq t\}$, for all $t \in \mathbb{R}$ and each $j = 1, \ldots, p_n$, where $1\{A\}$ is the indicator of $A$. Note that $n$ can be viewed as the number of microarrays while $p_n$ can be viewed as the number of genes.
mentioned in the introduction, our asymptotic interest focuses on what happens when \( n \) increases slowly while \( p_n \) increases rapidly.

We first establish, in section 2.1, uniform consistency of the marginal empirical distribution function estimator and also the uniformity of a Brownian bridge approximation to the standardized version of this estimator. These results are then used in section 2.2 to establish uniform consistency of the marginal sample means and uniform validity of marginal p-values based on the normal approximation to the t-test. The results are extended in section 2.3 for inference based on the marginal sample medians. Note that both the mean and median are functionals of the empirical distribution function. The mean is computationally simpler, but the median is more robust to data contamination.

**2.1 Consistency of the marginal empirical distribution functions.** The results of this section will form the basis for the results presented in sections 2.2 and 2.3. The two theorems of this section, theorems 1 and 2 below, are somewhat surprising, high dimensional extensions of two classical univariate results for empirical distribution functions: the celebrated Dvoretzky, Kiefer and Wolfowitz (1956) inequality as refined by Massart (1990) and the celebrated Komlós, Major and Tusnády (1976) Hungarian construction as refined by Bretagnolle and Massart (1989). The extensions utilize maximal inequalities based on Orlicz norms (see chapter 2.2 of van der Vaart and Wellner, 1996). For any real random variable \( Y \) and any \( d \geq 1 \), let \( \| Y \|_{\psi_d} \) denote the Orlicz norm for \( \psi_d(x) = e^{x^d} - 1 \), i.e., \( \| Y \|_{\psi_d} = \inf \left\{ C > 0 : \mathbb{E} \left[ e^{\left| Y \right|^d/C} - 1 \right] \leq 1 \right\} \).

Note that these norms increase with \( d \) (up to a constant depending only on \( d \)) and that \( \| \cdot \|_{\psi_1} \) dominates all \( L_p \) norms (up to a constant depending only on \( p \)). Also let \( \| \cdot \|_{\psi_\infty} \) be the uniform norm.

The first theorem we present yields simultaneous consistency of all the \( \hat{F}_{j(n)} \)'s for the corresponding \( F_{j(n)} \)’s:

**Theorem 1** There exists a universal constant \( 0 < c_0 < \infty \) such that, for all \( n, p_n \geq 2 \),

\[
\left\| \max_{1 \leq j \leq p_n} \left\| \hat{F}_{j(n)} - F_{j(n)} \right\|_{\psi_\infty} \right\|_{\psi_2} \leq c_0 \sqrt{\frac{\log p_n}{n}}. \tag{2}
\]
In particular, if $n \to \infty$ and $\log p_n/n \to 0$, then the left-hand-side of (2) goes to zero.

**Remark 1** One can show that the rate on the right-side of (2) is sharp, in the sense that there exist sequences of data sets, where $(\log p_n/n)^{-1/2} \max_{1 \leq j \leq p_n} \| \hat{F}_{j(n)} - F_{j(n)} \|_\infty \to c$, in probability, as $n \to \infty$, and where $0 < c < \infty$. In particular, the statement is true if the genes are all independent, $n, p_n \to \infty$ with $\log p_n = o(n)$, and $c = 1/2$.

The second theorem shows that the standardized empirical processes $\sqrt{n}(\hat{F}_{j(n)} - F_{j(n)})$ can be simultaneously approximated by Brownian bridges in a manner which preserves the original dependency structure in the data. This feature will be useful in studying FDR (see Benjamini and Hochberg, 1995) properties later on. To this end, let $\mathcal{F}_{j(n)}$ denote the smallest $\sigma$-field making all of $X_{1j(n)}, \ldots, X_{nj(n)}$ measurable, $1 \leq j \leq p_n$. Also let $\mathcal{F}_n$ be the smallest $\sigma$-field making all of $\mathcal{F}_{1(n)}, \ldots, \mathcal{F}_{p_n(n)}$ measurable.

**Theorem 2** There exists universal constants $0 < c_1, c_2 < \infty$ such that, for all $n, p_n \geq 2$,

\[
\left\| \max_{1 \leq j \leq p_n} \left\| \sqrt{n}(\hat{F}_{j(n)} - F_{j(n)}) - B_{j(n)}(F_{j(n)}) \right\|_\infty \right\|_{\psi_1} \leq \frac{c_1 \log n + c_2 \log p_n}{\sqrt{n}},
\]

for some stochastic processes $B_{1(n)}, \ldots, B_{p_n(n)}$ which are conditionally independent given $\mathcal{F}_n$ and for which each $B_{j(n)}$ is a standard Brownian bridge with conditional distribution given $\mathcal{F}_n$ depending only on $\mathcal{F}_{j(n)}$, $1 \leq j \leq p_n$.

2.2 Estimation of marginal sample means. Now we consider marginal inference based on the marginal sample mean. For each $1 \leq j \leq p_n$, assume for this section that the closure of the support of $F_{j(n)}$ is a compact interval $[a_{j(n)}, b_{j(n)}]$ with $a_{j(n)} \neq b_{j(n)}$, and that $F_{j(n)}$ has mean $\mu_{j(n)}$ and standard deviation $\sigma_{j(n)} > 0$. Let $\bar{X}_{j(n)}$ be the sample mean of $X_{1j(n)}, \ldots, X_{nj(n)}$. The following corollary yields simultaneous consistency of the marginal sample means:
Corollary 1 Under the conditions of theorem 1 and with the same constant \(c_0\), we have for all \(n, p_n \geq 2\),

\[
\max_{1 \leq j \leq \infty} |\bar{X}_{j(n)} - \mu_{j(n)}| \leq c_0 \sqrt{\frac{\log p_n}{n}} \max_{1 \leq j \leq p_n} |b_{j(n)} - a_{j(n)}|.
\]

Remark 2 Note that corollary 1 slightly extends the large \(p\) small \(n\) consistency results of van der Laan and Bryan (2001) by allowing the range of the support to increase with \(n\) provided it does not increase too rapidly.

Now assume that we wish to test the marginal null hypothesis \(H_{0j(n)}^{j(n)}: \mu_{j(n)} = \mu_{0j(n)}\) with the test statistic

\[
T_{j(n)} = \frac{\sqrt{n}(\bar{X}_{j(n)} - \mu_{0j(n)})}{\hat{\sigma}_{j(n)}},
\]

where \(\hat{\sigma}_{j(n)}\) is a location-invariant and consistent estimator of \(\sigma_{j(n)}\). To use FDR, we need to obtain uniformly consistent estimates of the p-values of these tests. One way to do this is with permutation methods. A computationally easier way is to just use \(\hat{\pi}_{j(n)} = 2\Phi(-|T_{j(n)}|)\), where \(\Phi\) is the distribution function for the standard normal. The conclusion of the following corollary is that this approach leads to uniformly consistent p-values under reasonable conditions:

Corollary 2 Let the constants \(c_1, c_2\) be as in theorem 2. Then, for all \(n, p_n \geq 2\), there exist standard normal random variables \(Z_{1(n)}, \ldots, Z_{p_n(n)}\) which are conditionally independent given \(\mathcal{F}_n\) and for which each \(Z_{j(n)}\) has conditional distribution given \(\mathcal{F}_n\) depending only on \(\mathcal{F}_{j(n)}\), \(1 \leq j \leq p_n\), such that

\[
\max_{1 \leq j \leq p_n} |\hat{\pi}_{j(n)} - \pi_{j(n)}| \leq \frac{c_1 \log n + c_2 \log p_n}{\sqrt{n}} \left( \max_{1 \leq j \leq p_n} \frac{|b_{j(n)} - a_{j(n)}|}{\sigma_{j(n)}} \right) + \frac{1}{2} \left( \max_{1 \leq j \leq n} \left( \frac{1}{\sigma_{j(n)}} \sigma_{j(n)} \right) \frac{1}{\sigma_{j(n)}} - 1 \right),
\]
where \( x \vee y \) denotes the maximum of \( x, y \) and

\[
\pi_j(n) = 2\Phi \left( -\frac{Z_j(n) + \sqrt{n}(\mu_j(n) - \mu_{0,j}(n))}{\sigma_j(n)} \right).
\]

In particular, if \( n \to \infty, \max_{1 \leq j \leq p_n} |\hat{\sigma}_j(n) - \sigma_j(n)|/(\sigma_j(n)\hat{\sigma}_j(n)) \to 0 \) in probability, and

\[
\frac{\log(n \vee p_n)}{\sqrt{n}} \times \max_{1 \leq j \leq p_n} \left| \frac{b_j(n) - a_j(n)}{\sigma_j(n)} \right| \to 0,
\]

then the left-hand-side of (6) \( \to 0 \) in probability.

**Remark 3** When \( |b_j(n) - a_j(n)|/\sigma_j(n) \) is bounded, condition (7) becomes \( \log^2 p_n/n = o(1) \).

**Remark 4** Now, suppose the indices \( J_n = \{1, \ldots, p_n\} \) are divided into two groups, \( J_{0n} \) and \( J_{1n} \), where \( H_{0j}^{(n)} \) holds for all \( j \in J_{0n} \) and where \( \delta_j^{(n)} = |\mu_j(n) - \mu_{0,j}(n)|/\sigma_j(n) > \tau \) for all \( j \in J_{1n} \), where \( \tau > 0 \). Then all of the \( \hat{\pi}_j(n) \)'s for \( j \in J_{0n} \) will simultaneously converge to uniform random variables with the same dependency structure inherent in the data (as per the discussion before theorem 2 above). Moreover, all of the \( \hat{\pi}_j(n) \) for \( j \in J_{1n} \) will simultaneously converge to 0. Thus the q-value approach to controlling FDR given in Storey, Taylor and Siegmund (2004) should work under their weak dependence conditions (7)–(9) (see also their theorem 5). A minor adjustment to this argument will also work for contiguous alternative hypotheses where the \( \sqrt{n}\delta_j(n) \) quantities converge to bounded constants.

### 2.3 Estimation of marginal sample medians.

Now we consider inference for the median. Assume that each \( F_{j(n)} \) has median \( \xi_{j(n)} \) and is continuous in a neighborhood of \( \xi_{j(n)} \) with density \( f_{j(n)} \). In this section, we do not require the support of \( F_{j(n)} \) to be compact. We do, however, assume that there exists \( \eta, \tau > 0 \) such that

\[
\min_{1 \leq j \leq p_n} \inf_{x : |x - \xi_{j(n)}| \leq \eta} f_{j(n)}(x) \geq \tau.
\]
Denote the sample median for $X_{1j(n)}, \ldots, X_{nj(n)}$ as $\hat{\xi}_{j(n)}$. More precisely, let $\hat{\xi}_{j(n)} = \inf\{x : \hat{F}_{j(n)}(x) \geq 1/2\}$. The following corollary gives simultaneous consistency of $\hat{\xi}_{j(n)}$:

**Corollary 3** Under condition (8) (for some $\eta, \tau > 0$) and the conditions of corollary 1, we have that

$$\max_{1 \leq j \leq p} |\hat{\xi}_{j(n)} - \xi_{j(n)}| = O_P\left(\frac{\log(n \vee p_n)}{n} + \sqrt{\frac{\log p_n}{n}}\right).$$  

Now assume that we wish to test the marginal null hypothesis $H_{0j(n)}^j : \xi_{j(n)} = \xi_{0j(n)}$ with the test statistics $U_{j(n)} = 2\sqrt{n} \hat{f}_{j(n)}(\hat{\xi}_{j(n)} - \xi_{0j(n)})$, where $\hat{f}_{j(n)}$ is a consistent estimator of $f_{j(n)}(\xi_{j(n)})$. As discussed in Kosorok (1999), this is a good choice of median test because it converges rapidly to its limiting Gaussian distribution and appears to have better moderate sample size performance compared to other median tests. As with the marginal mean test, we need consistent estimates of the p-values of these tests. We now study the consistency of the p-value estimates $\hat{\pi}'_{j(n)} = 2\Phi(-|U_{j(n)}|)$. We need some additional conditions. Assume there exists $\eta, \tau > 0$ and $M < \infty$ such that (8) holds and, moreover, that

$$\max_{1 \leq j \leq p} \sup_{x : |x - \xi_{j(n)}| \leq \eta} |f_{j(n)}(\xi_{j(n)} + u) - f_{j(n)}(\xi_{j(n)})| \leq M.$$  

and

$$\max_{1 \leq j \leq p} \sup_{\tau \leq \eta} \sup_{u : |u| \leq \epsilon} \frac{|f_{j(n)}(\xi_{j(n)} + u) - f_{j(n)}(\xi_{j(n)})|}{\epsilon^{1/2}} \leq M.$$  

We now have the following corollary:

**Corollary 4** Under conditions (8), (10) and (11), for some $\eta, \tau > 0$ and $M < \infty$, and provided both $\max_{1 \leq j \leq p} |\hat{f}_{j(n)} - f_{j(n)}(\xi_{j(n)})| = o_P(1)$ and $\log^3 p_n/n \to 0$ as
\( n \to \infty \), we have that

\[
\max_{1 \leq j \leq p_n} |\hat{\pi}'_j(n) - \pi'_j(n)| = o_P(1),
\]

where

\[
\pi'_j(n) = 2\Phi \left( -|Z_j(n) + 2\sqrt{\hat{f}_j(n)}(\xi_j(n))\xi_j(n) - \xi_{0,j}(n)| \right),
\]

and, for each \( n \geq 1 \), \( Z_{1(n)}, \ldots, Z_{p_n(n)} \) are standard normals conditionally independent given \( \mathcal{F}_n \) and for which each \( Z_j(n) \) has conditional distribution given \( \mathcal{F}_n \) depending only on \( \mathcal{F}_{j(n)} \), \( 1 \leq j \leq p_n \).

Now, for corollary 4 to be useful in conducting inference, we need simultaneously consistent estimators \( \hat{f}_j(n) \). One possibility is

\[
\hat{f}_j(n) = \frac{\hat{F}_j(n) \xi_j(n) + \hat{h}_j(n) - \hat{F}_j(n) \xi_j(n) - \hat{h}_j(n)}{2\hat{h}_j(n)},
\]

where the window widths \( \hat{h}_j(n) \) are allowed to depend on the data but must satisfy

\[
\max_{1 \leq j \leq p_n} \hat{h}_j^{-1}(n) \left( \frac{\log n \vee p_n}{n} + \sqrt{\frac{\log p_n}{n}} \right) = o_P(1).
\]

If, in addition to the conditions of corollary 4, we assume conditions (8) and (10) apply to the lower and upper quartiles of the distributions \( F_{j(n)} \), then \( \hat{h}_j(n) = 2\hat{I}_j(n)n^{-1/5} \), where \( \hat{I}_j(n) \) is the sample interquartile range based on \( \hat{F}_j(n) \), satisfies this requirement. This can be argued by first noting that \( \hat{I}_j(n) \) is asymptotically simultaneously bounded above and below and that

\[
n^{-1/5} \sqrt{\frac{\log p_n}{n}} = \sqrt{\frac{\log p_n}{n^{3/5}}} = \sqrt{\frac{\log p_n}{n^{1/3} n^{-4/15}}} \to 0.
\]
There are many other possibilities that will also work.

3. Marginal asymptotics for two-sample comparisons. The results of section 2 can be extended to two sample results, where we have two i.i.d. samples of vectors of length $p_n$, where $n = n_1 + n_2$, and where $n_k$ is the size of sample $k$, for $k = 1, 2$. Consistency results for estimating marginal distribution functions, marginal means and marginal medians follows essentially without modification from theorem 1 and corollaries 1 and 3. Our interest will therefore focus on the more challenging issue of testing whether the marginal means or medians are the same between the two samples. We use superscript $(k)$ to denote membership in group $k$, for $k = 1, 2$. In particular, $X_{i(n)}^{(k)} = (X_{i1(n)}^{(k)}, \ldots, X_{ip_n(n)}^{(k)})'$ is the $i$th observed vector in the $k$th group. In a similar manner, $F_{j(n)}^{(k)}$, $a_{j(n)}^{(k)} \neq b_{j(n)}^{(k)}$, $\mu_{j(n)}^{(k)}$, $\sigma_{j(n)}^{(k)} > 0$, $\xi_{j(n)}^{(k)}$ and $f_{j(n)}^{(k)}$, for $1 \leq j \leq p_n$, $k = 1, 2$, and all $n \geq 1$, are the two-sample versions of the corresponding one-sample quantities introduced in section 2. Also let $F^*_{j(n)} = \sigma (F_{j(n)}^{(1)}, F_{j(n)}^{(2)})$ and $F^*_n = \sigma (F^*_1(n), \ldots, F^*_p(n))$.

We first consider comparing the marginal means. Let $\bar{X}_{j(n)}^{(k)}$ be the sample mean of $X_{1j(n)}^{(k)}, \ldots, X_{nj(n)}^{(k)}$. Now assume that we wish to test the marginal null hypothesis $H_0^{(j(n))}: \mu_{j(n)}^{(1)} = \mu_{j(n)}^{(2)}$ with the test statistic

$$T_{j(n)}^* = \sqrt{\frac{n_1 n_2}{n_1 \left[ \hat{\sigma}_{j(n)}^{(2)} \right]^2 + n_2 \left[ \hat{\sigma}_{j(n)}^{(1)} \right]^2}} \left( \bar{X}_{j(n)}^{(1)} - \bar{X}_{j(n)}^{(2)} \right),$$

where $\hat{\sigma}_{j(n)}^{(k)}$ is a location-invariant and consistent estimator of $\sigma_{j(n)}^{(k)}$, $k = 1, 2$. The following corollary provides conditions under which p-values estimated by $\hat{\pi}_{j(n)}^* = 2\Phi \left( -|T_{j(n)}^*| \right)$ are uniformly consistent over all $1 \leq j \leq p_n$:

**Corollary 5** Let the constants $c_1, c_2$ be as in theorem 2. Then for all $n_1, n_2, p_n \geq 2$, there exist standard normal random variables $Z_{1(n)}^*, \ldots, Z_{p_n(n)}^*$ which are conditionally independent given $F^*_n$ and for which each $Z_{j(n)}^*$ has conditional distribution
given \( F_n^* \) depending only on \( F_{j(n)}^* \), \( 1 \leq j \leq p_n \), such that

\[
\max_{1 \leq j \leq p_n} \left| \hat{\pi}_{j(n)}^* - \pi_{j(n)}^* \right| \leq \sum_{k=1,2} \left[ \frac{c_1 \log n_k + c_2 \log p_n}{\sqrt{n_k}} \left( \max_{1 \leq j \leq p_n} \frac{|\hat{b}_{j(n)}^{(k)} - a_{j(n)}^{(k)}|}{\sigma_{j(n)}^{(k)}} \right) \right]
\]

\[
+ \frac{1}{2} \left( \max_{1 \leq j \leq n} \left( \hat{\sigma}_{j(n)}^{(k)} \right) \right) \left( \frac{1}{\hat{\sigma}_{j(n)}^{(k)}} - \frac{1}{\sigma_{j(n)}^{(k)}} \right) \right],
\]

where

\[
\pi_{j(n)}^* = 2\Phi \left( -Z_{j(n)}^* + \sqrt{\frac{n_1 n_2}{n_1 \left( \sigma_{j(n)}^{(2)} \right)^2 + n_2 \left( \sigma_{j(n)}^{(1)} \right)^2}} \left( \mu_{j(n)}^{(1)} - \mu_{j(n)}^{(2)} \right) \right).
\]

In particular, if \( n_k \to \infty \), \( \max_{1 \leq j \leq p_n} \left| \hat{\sigma}_{j(n)}^{(k)} - \sigma_{j(n)}^{(k)} \right| \to 0 \) in probability, and

\[
\frac{\log(n_k \vee p_n)}{\sqrt{n_k}} \times \max_{1 \leq j \leq p_n} \frac{|\hat{b}_{j(n)}^{(k)} - a_{j(n)}^{(k)}|}{\sigma_{j(n)}^{(k)}} \to 0,
\]

for \( k = 1, 2 \), then the left-hand-side of (16) \( \to 0 \) in probability.

We now consider comparing marginal medians. Assume that we wish to test the marginal null hypothesis \( H_0^{j(n)} : \xi_{j(n)}^{(1)} = \xi_{j(n)}^{(2)} \) with the test statistic

\[
U_{j(n)}^\# = 2 \sqrt{\frac{n_1 n_2}{n_1 \left( \hat{f}_{j(n)}^{(2)} \right)^2 + n_2 \left( \hat{f}_{j(n)}^{(1)} \right)^2}} \left( \xi_{j(n)}^{(1)} - \xi_{j(n)}^{(2)} \right),
\]

where \( \hat{f}_{j(n)}^{(k)} \) is consistent for \( f_{j(n)}^{(k)}(\xi_{j(n)}^{(k)}) \), \( k = 1, 2 \). The following corollary provides conditions under which p-values estimated by \( \hat{\pi}_{j(n)}^\# = 2\Phi \left( -|U_{j(n)}^\#| \right) \) are uniformly consistent over all \( 1 \leq j \leq p_n \):
Corollary 6 Assume that the one-sample conditions given in expressions \((8)\), \((10)\) and \((11)\), for all of the marginal distribution functions and densities in both samples, are satisfied for constants \(\eta, \tau > 0\) and \(0 < M < \infty\). Assume also that
\[
\max_{k=1,2; 1 \leq j \leq p_n} \left| \hat{f}^{(k)}_{j(n)} - f^{(k)}_{j(n)}(\xi^{(k)}_{j(n)}) \right| = o_p(1) \text{ and } \log^3 p_n/(n_1 \wedge n_2) \to 0 \text{ as } n \to \infty.
\]
Then
\[
\max_{1 \leq j \leq p_n} \left| \hat{\pi}^{\#}_{j(n)} - \pi^{\#}_{j(n)} \right| = o_p(1),
\]
where
\[
\pi^{\#}_{j(n)} = 2\Phi \left( -Z_{j(n)}^* + 2 \sqrt{\frac{n_1 n_2}{n_1 f^{(2)}_{j(n)}(\xi^{(2)}_{j(n)})} + \frac{n_2}{n_2 f^{(1)}_{j(n)}(\xi^{(1)}_{j(n)})}} (\xi^{(1)}_{j(n)} - \xi^{(2)}_{j(n)}) \right),
\]
and, for each \(n \geq 1\), \(Z^*_{1(n)}, \ldots, Z^*_{p_n(n)}\) are standard normals conditionally independent given \(F_n^*\) and for which each \(Z^*_{j(n)}\) has conditional distribution given \(F_n^*\) depending only on \(F_{j(n)}^*\), \(1 \leq j \leq p_n\).

4. Distribution free statistics. When the distribution of the test statistic under the null hypothesis does not depend on the distribution function, results stronger than those presented in sections 2 and 3 are possible for marginal p-value consistency. Consider first the one-sample setting, and assume that the distributions \(F_{j(n)}\) are all continuous and symmetric around their respective medians. Suppose we are interested in marginal testing of \(H_0^{j(n)}: \xi_{j(n)} = 0\) using the signed rank test \(T_{j(n)}\) studied in section 3 of Kosorok and Ma (2005). Define
\[
V_{j(n)} = \frac{T_{j(n)} - (n^2 + n)/4}{\sqrt{(3n^3 + 2n^2 + n)/24}}.
\]
Note that the distribution of \(V_{j(n)}\) does not depend on \(F_{j(n)}\) under \(H_0^{j(n)}\). Let \(\Phi_n\) be the exact distribution of \(V_{j(n)}\) under \(H_0^{j(n)}\). It is easy to verify that \(\Phi_n\) converges
uniformly to \( \Phi \). Hence
\[
\max_{1 \leq j \leq p_n} |2\Phi_n(-|V_{j(n)}|) - 2\Phi(-|V_{j(n)}|)| \to 0,
\]
regardless of how fast \( p_n \) grows. Thus the normal approximation is simultaneously consistent for the true p-values when \( n \to \infty \), without any constraints on \( p_n \).

The key feature that makes this work is that the p-values depend only on the correctness of the probability calculation under the null hypothesis. P-value computations do not require knowledge of the distribution under alternatives. The only possibly unnatural assumption required for the above signed-rank test is symmetry about the median. An alternative statistic is the sign test. Under the null hypothesis that the median is zero, the sign test is Bernoulli with probability \( 1/2 \). As with the signed-rank test, the standardized sign test under the null converges to a normal limit. A disadvantage of the sign test is that the range of possible values is limited, resulting in a granular distribution which converges somewhat slowly to the normal limit.

Similar reasoning applies to distribution-free two-sample test statistics. Interestingly, there appears to be a larger variety of useful tests to choose from which do not require specification of the distribution function than there are in the one-sample setting. Suppose we are interested in marginal testing of \( H_0^{j(n)} : F_{j(n)}^{(1)} = F_{j(n)}^{(2)} \), and we assume that the \( F_{j(n)}^{(k)} \) are continuous for all \( 1 \leq j \leq p_n \) and \( k = 1, 2 \). Let
\[
\hat{F}_{j(n)}^{(k)}(t) = n_k^{-1} \sum_{i=1}^{n_k} \mathbf{1}\{X_{ij(n)}^{(k)} \leq t\}, \quad \text{for } k = 1, 2; \quad \hat{F}_{j(n)}^{(0)} = n^{-1} \left[ n_1 \hat{F}_{j(n)}^{(1)} + n_2 \hat{F}_{j(n)}^{(2)} \right];
\]
and \( \hat{G}_{j(n)} = \sqrt{n_1n_2/n} \left( \hat{F}_{j(n)}^{(1)} - \hat{F}_{j(n)}^{(2)} \right) \). We now consider several statistics which are invariant under monotone transformations of the data:

1. The two-sample Wilcoxon rank sum test \( \tilde{T}_{j(n)}^{*1} = \sqrt{12} \int_{\mathbb{R}} \hat{G}_{j(n)}(s) d\hat{F}_{j(n)}^{(0)}(s) \);
2. The two-sample Kolmogorov-Smirnov test \( \tilde{T}_{j(n)}^{*2} = \sup_{t \in \mathbb{R}} |\hat{G}_{j(n)}(t)| \);
3. The two-sample Cramér-von Mises test \( \tilde{T}_{j(n)}^{*3} = \int_{\mathbb{R}} \hat{G}_{j(n)}^{2}(s) d\hat{F}_{j(n)}^{(0)}(s). \)

Fix \( j \in \{1, \ldots, p_n\} \) and assume \( H_0^{j(n)} \) holds. All three of these statistics are now invariant under the monotone transformation \( t \mapsto F_{j(n)}^{(0)}(t) \), where \( F_{j(n)}^{(0)} \equiv F_{j(n)}^{(1)} = \)

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Thus, without loss of generality, we can assume the data are i.i.d. uniform \([0, 1]\). For \(m = 1, 2, 3\), let \(K_n^{*m}\) be the corresponding cumulative distribution function for the statistic \(\tilde{T}_{j(n)}^{*m}\) under this uniformity assumption (note that it does not depend on \(j\) because of the invariance), and let \(K_0^{*m}\) be the limiting cumulative distribution function. Suppose that we compute approximate p-values for the three statistics as follows: 
\[\hat{\pi}_{j(n)}^{*1} = 2\Phi\left(-\frac{1}{\sqrt{2}}\tilde{T}_{j(n)}^{*1}\right),\] 
\[\hat{\pi}_{j(n)}^{*m} = 1 - K_0^{*m}\left(\tilde{T}_{j(n)}^{*m}\right),\] 
for \(m = 2, 3\). Because it can be shown that \(K_0^{*m}\) is continuous for all \(m = 1, 2, 3\), the convergence of \(K_n^{*m}\) to \(K_0^{*m}\) is uniform. Thus, even after we drop the \(H_{j(n)}^2\) assumption, the approximate p-values based on \(K_n^{*m}\) are simultaneously consistent for the true p-values, as \(n \to \infty\), without constraining \(p_n\).

The following lemma yields the form of \(K_0^{*m}\), for \(m = 1, 2, 3\). The results are essentially classical, but they are included here for completeness:

**Lemma 1** For \(m = 1, 2, 3\), \(K_n^{*m}\) converges uniformly to \(K_0^{*m}\), as \(n_1 \land n_2 \to \infty\), where

- \(K_0^{*1} = \Phi;\)

- For \(t > 0\), \(K_0^{*2}(t) = 1 - 2\sum_{l=1}^{\infty}(-1)^l e^{-2l^2t^2}\) is the distribution of the supremum in absolute value of a standard Brownian bridge;

- \(K_0^{*3}\) is the distribution of \(\pi^{-2} \sum_{l=1}^{\infty} t^{-2} \tilde{Z}_l^2\), where \(\tilde{Z}_1, \tilde{Z}_2, \ldots\) are i.i.d. standard normals.

5. **Impact of microarray normalization.** In this section, we consider the affect of normalization on the theory presented in sections 2–4. For the simple normalization model \((\Pi)\), this will require the \(h_i\)s to be uniformly consistent at the rate \(O_P(\sqrt{n \log n})\). This requirement seems reasonable for certain estimation methods, including the method described in FPH. In this method, data across all genes within each array are utilized for estimating the \(h_i\)s. Since the number of genes \(p_n\) usually increases nearly exponentially relative to the number of microarrays, the number of observations available for estimating the \(h_i\)s is many orders of magnitude higher than \(n\), even after taking into account dependencies within arrays and the fact that the number of arrays is increasing in \(n\). For this particular facet of our
problem, the large number of genes actually works in our favor. A variant of this argument can also be found in Kosorok and Ma (2005).

Consider first the one-sample setting of section 2. Let \( \tilde{X}_{i(n)} = (\tilde{X}_{i1(n)}, \ldots, \tilde{X}_{ip_n(n)})' \) be an approximation of the “true data” \( X_{i(n)}, 1 \leq i \leq n \), and define

\[
\hat{\varepsilon}_n = \max_{1 \leq j \leq p_n; 1 \leq i \leq n} |\tilde{X}_{ij(n)} - X_{ij(n)}|.
\]

With proper, partially consistent normalization, the true gene effects \( \{X_{ij(n)}, 1 \leq j \leq p_n, 1 \leq i \leq n\} \) should be uniformly consistently estimated by the residuals from the normalization \( \{\tilde{X}_{ij(n)}, 1 \leq j \leq p_n, 1 \leq i \leq n\} \). In other words \( \hat{\varepsilon}_n = o_P(1) \). The essence of our arguments involves an assessment of how well \( \tilde{F}_j(n)(t) \equiv n^{-1} \sum_{i=1}^n 1\{\tilde{X}_{ij(n)} \leq t\} \) approximates \( \hat{F}_j(n)(t) \) uniformly in \( t \). We need the following strengthening of condition (10):

\[
\limsup_{n \to \infty} \max_{1 \leq j \leq p_n} \sup_{t \in \mathbb{R}} f_{j(n)}(t) \leq \tilde{M},
\]

for some \( \tilde{M} < \infty \). We now have the following theorem, the proof of which involves a precise bound on the modulus of continuity of Brownian motion (see lemma 2 in section 8 below):

**Theorem 3** Assume condition (21) holds for some \( \tilde{M} < \infty \). Then the following are true:

(i) If \( \log p_n/n = o(1) \) and \( \hat{\varepsilon}_n = o_P(1) \), then

\[
\max_{1 \leq j \leq p_n} \left\| \tilde{F}_j(n) - \hat{F}_j(n) \right\|_{\infty} = o_P(1);
\]

(ii) If, in addition, \( \log^2 p_n/n = o(1) \) and \( \sqrt{n} (\log n) \hat{\varepsilon}_n = O_P(1) \), then also

\[
\max_{1 \leq j \leq p_n} \left\| \tilde{F}_j(n) - \hat{F}_j(n) \right\|_{\infty} = o_P(n^{-1/2}).
\]
Remark 5 Note that the one-sample signed rank test $\tilde{T}_j(n)$ can be written as a normalization of $\sqrt{n} \int_\mathbb{R} \left[ \tilde{F}_j(n)(u) - \tilde{F}_j(n)(-u) \right] d\tilde{F}_j(n)(u)$, and the one-sample sign test can be written as a normalization of $\sqrt{n} \int_\mathbb{R} \text{sign}(u) d\tilde{F}_j(n)(u)$. Thus part (ii) of theorem 3 allows us to replace $\tilde{F}_j(n)$ with $\tilde{F}_j(n)$ in both of these statistics without destroying the simultaneous consistency over $1 \leq j \leq p_n$ established in section 4 of the normal approximation for the true p-values based on the true data.

Theorem 3 can also be used to verify that the asymptotic results for the one-sample mean and median tests of sections 2 and 3 can be similarly extended for the approximate data $\tilde{X}_1(n), \ldots, \tilde{X}_n(n)$. For $j = 1, \ldots, p_n$, let $\tilde{X}_j(n)$ be the sample mean of $\tilde{X}_{1j}(n), \ldots, \tilde{X}_{nj}(n)$, and define the approximate sample median $\tilde{\xi}_j(n) = \inf \{ r : \tilde{F}_j(n)(r) \geq 1/2 \}$. The following corollary yields consistency of these estimators:

**Corollary 7** Assume the conditions of theorem 3 part (i), hold. Then

(i) $\max_{1 \leq j \leq p_n} \left\| \tilde{F}_j(n) - F_j(n) \right\|_\infty = o_P(1)$;

(ii) Provided $\limsup_{n \to \infty} \max_{1 \leq j \leq p_n} b_{j(n)} - a_{j(n)} < \infty$, $\max_{1 \leq j \leq p_n} |\tilde{X}_j(n) - \mu_{j(n)}| = o_P(1)$;

(iii) $\max_{1 \leq j \leq p_n} |\tilde{\xi}_j(n) - \xi_{j(n)}| = o_P(1)$.

The following corollary strengthens result (ii) of corollary 7 above and yields consistency of the p-values of one-sample tests based on the approximate data:

**Corollary 8** Assume the conditions of theorem 3 part (ii), hold. Then the following results are true under the given conditions:

(i) Provided $\limsup_{n \to \infty} \max_{1 \leq j \leq p_n} n^{1/4} |b_{j(n)} - a_{j(n)}| < \infty$, $\max_{1 \leq j \leq p_n} |\tilde{X}_j(n) - \mu_{j(n)}| = o_P(1)$.

(ii) Suppose that the conditions of corollary 7 hold, except that $\tilde{X}_j(n)$ is used instead of $\tilde{X}_j(n)$ and that all other estimated quantities are based on $\tilde{F}_j(n)$ rather than on $\tilde{F}_j(n)$, for $j = 1, \ldots, p_n$. Then, provided

$$\limsup_{n \to \infty} \max_{1 \leq j \leq p_n} \left\{ \frac{|b_{j(n)} - a_{j(n)}|}{\sigma_{j(n)}} \vee 1 \right\} < \infty$$
and \( \max_{1 \leq j \leq p_n} |\hat{a}_{j(n)} - \sigma_{j(n)}| / (\sigma_{j(n)} \hat{a}_{j(n)}) \to 0, \max_{1 \leq j \leq p_n} |\hat{\pi}_{j(n)} - \pi_{j(n)}| = o_P(1), \) for the filtrations \( F_n \) and \( F_{j(n)} \), \( 1 \leq j \leq p_n \), based on the true data.

(iii) Suppose that the conditions of corollary \( 4 \) hold, except that \( \hat{\xi}_{j(n)} \) is used instead of \( \hat{\xi}_{j(n)} \) and that all other estimated quantities are based on \( \hat{F}_{j(n)} \) rather than on \( \tilde{F}_{j(n)} \), for \( j = 1, \ldots, p_n \). Then the conclusions of corollary \( 4 \) still hold for the filtrations \( F_n \) and \( F_{j(n)} \), \( 1 \leq j \leq p_n \), based on the true data.

**Remark 6** Parts (ii) and (iii) of corollary \( 4 \) tell us that we can construct valid mean and median based hypothesis tests from suitably normalized data, and that any dependencies beyond the original dependency structure induced by the approximation vanish asymptotically. Thus the arguments given in remark \( 4 \) regarding the validity of the q-value approach for controlling FDR still hold after normalization.

The extension of these results to the two-sample setting is straightforward. As done in section 4, we will use superscript \((k)\) to denote membership in group \( k \), for \( k = 1, 2 \). Let \( \tilde{F}_{j(n)}^{(k)} \) be the empirical distribution of the approximate data sample \( \tilde{X}_{1j(n)}^{(k)}, \ldots, \tilde{X}_{nj(n)}^{(k)} \); \( \xi_{j(n)}^{(k)} = \inf \{ r : \tilde{F}_{j(n)}^{(k)}(r) \geq 1/2 \} \); \( \epsilon_n^{(k)} \) be the maximum error between the approximate and true data for group \( k \); and redefine \( \epsilon_n = \epsilon_n^{(1)} \vee \epsilon_n^{(2)} \). Also let \( \hat{T}_{j(n)}^{(m)} \) be the version of \( \hat{T}_{j(n)} \) with \( \tilde{F}_{j(n)}^{(k)} \) replacing \( \tilde{F}_{j(n)} \), for \( k = 1, 2 \) and \( m = 1, 2, 3 \). The following corollary gives the main two-sample approximation results:

**Corollary 9** Assume \( n_1 \wedge n_2 \to \infty \); \( \limsup_{p_n \to \infty} \max_{1 \leq k \leq 2} \max_{1 \leq j \leq p_n} \| f_{j(n)}^{(k)} \|_\infty \leq \tilde{M} \), for some \( \tilde{M} < \infty \); \( \log^2 p_n / (n_1 \wedge n_2) = o(1) \); and \( \sqrt{n} (\log n) \epsilon_n = O_P(1) \). Then the following are true under the given conditions:

(i) Suppose that the conditions of corollary \( 4 \) hold, except the sample means are based on the approximate data and all other estimated quantities are based on \( \tilde{F}_{j(n)}^{(k)} \) rather than on \( \tilde{F}_{j(n)}^{(k)} \), for \( j = 1, \ldots, p_n \) and \( k = 1, 2 \). Then, provided

\[
\limsup_{n \to \infty} \max_{k=1,2} \max_{1 \leq j \leq p_n} \frac{\{|\hat{b}_{j(n)}^{(k)} - a_{j(n)}^{(k)}| \vee 1\}}{\sigma_{j(n)}^{(k)}} < \infty
\]
\[
\text{and } \max_{k=1,2} \max_{1 \leq j \leq p_n} \left| \frac{\hat{\sigma}^{(k)}_{j(n)} - \sigma^{(k)}_{j(n)}}{\left(\sigma^{(k)}_{j(n)} \hat{\sigma}^{(k)}_{j(n)}\right)} \right| \to 0, \\
\max_{1 \leq j \leq p_n} \left| \frac{\hat{\pi}^*_{j(n)} - \pi^*_{j(n)}}{\sigma^{(k)}_{j(n)}} \right| = o_P(1),
\]

for the filtrations \(F_n\) and \(F_{j(n)}\), \(1 \leq j \leq p_n\), based on the true data.

(ii) Suppose that the conditions of corollary \(6\) hold, except that \(\hat{\xi}^{(k)}_{j(n)}\) is used instead of \(\tilde{\xi}^{(k)}_{j(n)}\) and that all other estimated quantities are based on \(\hat{F}^{(k)}_{j(n)}\) rather than on \(\tilde{F}^{(k)}_{j(n)}\), for \(j = 1, \ldots, p_n\) and \(k = 1, 2\). Then the conclusions of corollary \(6\) still hold for the filtrations \(F_n\) and \(F_{j(n)}\), \(1 \leq j \leq p_n\), based on the true data.

(iii) \[\max_{1 \leq j \leq p_n} \left| \hat{T}^{*m}_{j(n)} - \tilde{T}^{*m}_{j(n)} \right| = o_P(1), \text{ for } m = 1, 2, 3.\] Thus the approximate p-values based on the approximate data for the three distribution-free two-sample tests given in section 4 are uniformly consistent for the true p-values based on the true data.

6. Numerical studies.

6.1 One-sample simulation study. We used a small simulation study to assess the finite sample performance of the following one-sample methodologies: (1) the mean based comparison of section 2.2, (2) the median based comparison of section 2.3 and (3) the signed rank test of section 4. We set the number of genes to \(p = 2000\) and the number of arrays to \(n = 20, 50\). Let \(Z_{i1}, Z_{i2}, \ldots, i = 1, \ldots, n\), be a sequence of i.i.d. standard normal random variables. We generated simulated data using the following three models:

Model 1: \(X_{ij} = H(Z_{ij})\) for \(i = 1, \ldots, p\);

Model 2: \(X_{ij} = H\left(\sum_{l=(j-1)\times m+1}^{(j-1)\times m+k} Z_l / \sqrt{k}\right)\) with \(k = 10, m = 7\);

Model 3: Same as Model 2, but with \(k = 10, m = 3\).

In the above, \(H = 2\Phi - 1\), where \(\Phi\) is the cumulative distribution for the standard normal. This yields a marginal \(\text{unif}[−1,1]\) distribution for all three models. The genes in model 1 are i.i.d., while in model 2 there is strong dependence and in model 3 weak dependence between genes. We assume the first 40 genes have non-zero means, denoted as \(\beta\) and generated from \(\text{unif}[−2, 2]\). For each approach, marginal p-values are computed based on the asymptotic results for one-sample tests given
in sections 2 and 4. For the median approach, density estimation is based on the interquartile range band-width kernel described in the last paragraph of section 2.3. We employ standard FDR techniques with expected FDR $E(FDR) = 0.2$. The marginal p-values are ranked, resulting in the ordered p-values $\pi(1) \leq \pi(2) \leq \ldots \leq \pi(p)$. Let $\tilde{g}$ be the largest $g$ such that $\pi(g) \leq g/p \times q$, where $q$ is the target FDR (for the simulations, $q = 0.2$). Genes corresponding to $\pi(1), \ldots, \pi(\tilde{g})$ are identified as significantly differentially expressed.

Simulation results based on 100 replicates per scenario are shown in Table 1. We can see that as the sample size increases, the performances of all three approaches generally improve. When the sample size is small, the mean based approach can effectively identify differentially expressed genes, but with high false positive rates. Empirical FDRs for the rank approach are quite low. The rank based approach misses quite a few true positives. When the sample size is large, the median approach and the rank approach perform much better than the mean based approach, with less false positives while still being able to identify true positives. The presence of correlation appears to have very little impact on the performance.

6.2 Two-sample simulation study. Since the affect of dependence between genes in the simulation study of section 6.1 was minimal, we decided to restrict our focus on the i.i.d. gene setting for the two-sample simulations. We set the number of genes to $p = 2000$ and numbers of arrays (sample sizes) to $n_1 = n_2 = 10, 30, 60$. The model we explore is Model 4: $X_{ij}^{(k)} \sim \text{unif}[-1, 1]$, $i = 1, \ldots, k$, $j = 1, \ldots, p$, and $k = 1, 2$. For this data, we apply the mean approach, the median approach, the Wilcoxon test and the Kolmogorov-Smirnov test to the two-sample comparison of $X_{ij}^{(1)} + \beta$ versus $X_{ij}^{(2)}$, where $\beta$ is generated as in section 6.1 for the first 40 genes of each array. Summary statistics for $E(FDR) = 0.2$ and 100 replicates are shown in Table 2. Similar conclusions as in section 6.1 on the effects of sample size and gene distribution can be made. We especially notice that when the sample size is small, the mean based approach appears to be the only one that can identify a significant number of true positives. The false positive rates are smaller than the target for the median, Wilcoxon and Kolmogorov-Smirnov (KS) approaches. The mismatch between the empirical FDR with the target FDR can be serious for the mean approach, especially when the sample size is small.
Based on other numerical studies (not presented), it appears that part of the convergence difficulties with the nonparametric approaches (in both the one and two sample settings) are due to the small number of distinct possible values these statistics can have. It is unclear how to solve this problem for the nonparametric one-sample tests, but it appears that the two-sample tests can be improved by replacing $\hat{G}_{j(n)}$ with $\tilde{G}_{j(n)} = \sqrt{n_1 n_2 / n} \left[ \hat{F}_{j(n)}^{(1)} - \hat{F}_{j(n)}^{(2)} \right]$, where $\hat{F}_{j(n)}^{(2)} = (n_2 / (n_2 + 1)) \hat{F}_{j(n)}^{(2)}$. This increases the number of possible values of the statistic, and preliminary simulation studies (also not presented) indicate that the rate of convergence for smaller sample sizes is improved. Thus we recommend that this modification be considered whenever $n_1 = n_2$. Note that the modification does not affect the asymptotics since

$$\sqrt{n_1 n_2 / n} \left| 1 - \frac{n_2}{n_2 + 1} \right| \leq \frac{1}{\sqrt{n_2}}.$$ 

6.3 Estrogen data. These datasets were first presented by West et al. (2001) and Spang et al. (2001). Their common expression matrix monitors 7129 genes in 49 breast tumor samples. The data were obtained by applying the Affymetrix gene chip technology. The response describes the lymph nodal (LN) status, which is an indicator for the metastatic spread of the tumor, an important risk factor for disease outcome. 25 samples are positive (LN+) and 24 samples are negative (LN-). The goal is to identify genes differentially expressed between positive and negative samples from the 3332 genes passing the first step of processing described in Dudoit, Fridlyand, and Speed (2002). A base 2 logarithmic transformation of the gene expressions is first applied.

We set the target FDR to 0.1 and apply the standard FDR method with the four two-sample comparison approaches: 445 (mean), 261 (median), 423 (Wilcoxon) and 211 (KS) genes are identified, respectively. The mean based approach and the Wilcoxon test identify significantly more genes than the median approach and the KS test. This pattern was also demonstrated in Table 2 (for sample size $n_1 = n_2 = 30$). It is unclear what causes these differences. However, the overlaps of genes identified by the different approaches are substantial. For example, there are 196 common genes between the mean approach and the median approach. In Figure 1 we show
scatter plots of p-values from the different approaches. The rank correlation coefficients show substantial similarities among different approaches. Note the banded pattern in the plots involving the KS statistic. This is a consequence of the low number of distinct possible values this statistic can have as was discussed in section 6.2 above.

7. Discussion. The main results of this paper are that marginal (gene specific) estimates and asymptotic-based p-values are uniformly consistent in microarray experiments with \( n \) replications—regardless of the dependencies between genes—provided the number of genes \( p_n \) satisfies \( \log p_n = o(n) \), \( \log p_n = o(n^{1/2}) \) or \( \log p_n = o(n^{1/3}) \), depending on the desired task. In other words, the number of genes is allowed to increase almost exponentially fast relative to the number of arrays. This seems to be a realistic asymptotic regime for microarray studies. These results also hold true for two-sample comparisons. Moreover, the results continue to hold even after normalization, provided the normalization process is sufficiently accurate.

We note that the simulation and data analyses seem to support the theoretical results of the paper, although some test procedures appear to work better than others. We also acknowledge that a number of important issues, such as the affect of marginal distribution on the asymptotics and the affect of normalization, were not evaluated in the limited simulation studies presented in section 6. A refined and more thorough simulation study that addresses these points is beyond the scope of the current paper but is worth pursuing in the future.

A theoretical limitation of the present study is that the asymptotics developed are not yet accurate enough to provide precise guidelines on sample size for specific microarray experiments. The development of such guidelines is worthwhile to pursue as a future topic, but it most likely would require at least some assumptions on the dependencies between genes. Such assumptions are out of place in the present paper since a strength of the paper is the absence of assumptions on gene interdependence. It is because of this generality that we believe the results of this paper should be a useful point of departure for future, more refined asymptotic analyses of microarray experiments.
8. Proofs.

Proof of theorem 1. Define \( V_{j(n)} \equiv \sqrt{n} \| \hat{F}_{j(n)} - F_{j(n)} \|_{\infty} \), and note that by theorem 2 below combined with lemma 2.2.1 of van der Vaart and Wellner (1996) (abbreviated VW hereafter), \( \| V_{j(n)} \|_{\psi_2} \leq 3/2 \) for all \( 1 \leq j \leq p_n \). Now, by lemma 2.2.2 of VW combined with the fact that \( \limsup_{x,y \to \infty} \psi_2(x)\psi_2(y)/\psi_2(xy) = 0 \), we have that there exists a universal constant \( c_* < \infty \) such that

\[
\max_{1 \leq j \leq p_n} V_{j(n)} \leq c_0 \sqrt{\log(1 + p_n)} \leq c_0 \sqrt{3/2} \]

for all \( n \geq 1 \). The desired result now follows for the constant \( c_0 = \sqrt{6} c_* \), since \( \log(k + 1) \leq 2 \log k \) for any \( k \geq 2 \).

Theorem 4 Let \( Y_1, \ldots, Y_n \) be an i.i.d. sample of real random variables with distribution \( G \) (not necessarily continuous), and let \( \hat{G}_n \) be the corresponding empirical distribution function. Then

\[
P \left( \sup_{t \in \mathbb{R}} \sqrt{n} \left| \hat{G}_n(t) - G(t) \right| > x \right) \leq 2 e^{-2x^2},
\]

for all \( x \geq 0 \).

Proof. This is the celebrated result of Dvoretzky, Kiefer and Wolfowitz (1956), given in their lemma 2, as refined by Massart (1990) in his corollary 1. We omit the proof of their result but note that their result applies to the special case where \( G \) is continuous. We now show that it also applies when \( G \) may be discontinuous. Without loss of generality, assume that \( G \) has discontinuities, and let \( T_1, \ldots, T_m \) be the locations of the discontinuities of \( G \), where \( m \) may be infinity. Note that the number of discontinuities can be at most countable. Let \( r_1, \ldots, r_m \) be the jump sizes of \( G \) at \( T_1, \ldots, T_m \). Now let \( U_1, \ldots, U_n \) be i.i.d. uniform random variables independent of the \( Y_1, \ldots, Y_n \), and define new random variables

\[
Z_i = Y_i + \sum_{j=1}^{m} r_j \left[ 1 \{ T_j < Y_i \} + 1 \{ T_j = Y_i \} U_i \right], \quad 1 \leq i \leq n.
\]

Define also the transformation \( t \mapsto R(t) = t + \sum_{j=1}^{m} r_j 1 \{ T_j \leq t \} \); let \( \hat{H}_n^* \) be the empirical distribution of \( Z_1, \ldots, Z_n \); and let \( H \) be the distribution of \( Z_1 \). It is not hard to verify that

\[
\sup_{t \in \mathbb{R}} \left| \hat{G}_n(t) - G(t) \right| = \sup_{t \in \mathbb{R}} \left| \hat{H}_n^*(R(t)) - H(R(t)) \right|
\]

\[
\leq \sup_{s \in \mathbb{R}} \left| \hat{H}_n(s) - H(s) \right|,
\]

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and the desired result now follows since $H$ is continuous. □

Proof of theorem 2 Let $U_{ij(n)}$, $i = 0, \ldots, n$ and $j = 1, \ldots, p_n$, be independent uniform random variables. Then, by theorem 3 below, there exist Brownian bridges $B_{1(n)}, \ldots, B_{p_n(n)}$, where, for each $1 \leq j \leq p_n$, $B_{j(n)}$ depends only on $X_{1j(n)}, \ldots, X_{nj(n)}$ and $U_{0j(n)}, \ldots, U_{nj(n)}$ and

$$P\left(\sqrt{n}\|\sqrt{n}(\hat{F}_{j(n)} - F_{j(n)}) - B_{j(n)}(F_{j(n)})\|_\infty > x + 12 \log n\right) \leq 2e^{-x/6},$$

(22)

for all $x \geq 0$ and all $n \geq 2$. Now define

$$U_{j(n)} = \left(\frac{\sqrt{n}}{\log n}\|\sqrt{n}(\hat{F}_{j(n)} - F_{j(n)}) - B_{j(n)}(F_{j(n)})\|_\infty - 12\right)^+,$$

where $u^+$ is the positive part of $u$. By lemma 2.2.1 of VW, expression (22) implies that $\|U_{j(n)}\|_{\psi_1} \leq 18/\log n$. Reapplying the result that $\log(k + 1) \leq 2\log k$ for any $k \geq 2$, we now have, by the fact that $\lim sup_{x,y \to \infty} \psi_1(x)\psi_1(y)/\psi_1(xy) = 0$ combined with lemma 2.2.2 of VW, that there exists a universal constant $0 < c_2 < \infty$ for which

$$\left\|\max_{1 \leq j \leq p_n} U_{j(n)}\right\|_{\psi_1} \leq \frac{c_2 \log p_n}{\log n}.$$

Now (23) follows, for $c_1 = 12$, from the definition of $U_{j(n)}$. □

Theorem 5 For $n \geq 2$, let $Y_1, \ldots, Y_n$ be i.i.d. real random variables with distribution $G$ (not necessarily continuous), and let $U_0, \ldots, U_n$ be independent uniform random variables independent of $Y_1, \ldots, Y_n$. Then there exists a standard Brownian motion $B$ depending only on $Y_1, \ldots, Y_n$ and $U_0, \ldots, U_n$ such that, for all $x \geq 0$,

$$P\left(\sqrt{n}\|\sqrt{n}(\hat{G}_n - G) - B(G)\|_\infty > x + 12 \log n\right) \leq 2e^{-x/6},$$

(23)

where $\hat{G}_n$ is the empirical distribution of $Y_1, \ldots, Y_n$.

Proof. We will apply the same method for handling the discontinuities of $G$
as used in the proof of theorem 4. Let \( m \geq 0, T_1, \ldots, T_m, \) and \( r_1, \ldots, r_m \) be as defined in the proof of theorem 4. Similarly define \( Z_1, \ldots, Z_m, R, \hat{H}_n \) and \( H \), except that we will utilize the uniform random variables \( U_1, \ldots, U_n \) given in the statement of theorem 5. By the continuity of \( H \) as established in the proof of theorem 4, \( H(Z_1) \) is now uniformly distributed. Thus, by the Hungarian construction theorem (theorem 1) of Bretagnolle and Massart (1989), there exists a Brownian bridge \( B \) depending only on \( Z_1, \ldots, Z_n \) and \( U_0 \) such that

\[
P \left( \sqrt{n} \left\| \sqrt{n}(\hat{H}_n - H) - B(H) \right\|_\infty > x + 12 \log n \right) \leq 2e^{-x/6},
\]

for all \( x \geq 0 \). The desired result now follows since

\[
\sup_{t \in \mathbb{R}} |\sqrt{n}(\hat{G}_n(t) - G(t)) - B(G(t))| = \sup_{t \in \mathbb{R}} |\sqrt{n}(\hat{H}_n(R(t)) - H(R(t))) - B(H(R(t)))| \\
\leq \sup_{s \in \mathbb{R}} |\sqrt{n}(\hat{H}_n(s) - H(s)) - B(H(s))|.
\]

Proof of corollary 7. The result is a consequence of theorem 4 via the following integration by parts identity:

\[
\int_{[a_j(n), b_j(n)]} x \left[ d\hat{F}_{j(n)}(x) - dF_{j(n)}(x) \right] = - \int_{[a_j(n), b_j(n)]} \left[ \hat{F}_{j(n)}(x) - F_{j(n)}(x) \right] dx.
\]

Proof of corollary 2. Note that for any \( x \in \mathbb{R} \) and any \( y > 0 \),

\[
|\Phi(xy) - \Phi(x)| \leq \sup_{1 \leq y \leq 1+v} |x|\phi(xu)|y-1| \\
\leq 0.25 \times \sup_{1 \leq y \leq 1+v} \left| \frac{y-1}{u} \right| \\
\leq 0.25 \times \left| 1 - y \right| \vee \left| 1 - \frac{1}{y} \right|.
\]
The constant 0.25 comes from the fact that $\sup_{u > 0} u \phi(u) \leq (2\pi e)^{-1/2} \leq 0.25$. Thus

$$
(25) \quad \max_{1 \leq j \leq p_n} \left| \hat{\pi}_j(n) - \hat{\pi}_j^*(n) \right| \leq \frac{1}{2} \left( \max_{1 \leq j \leq n} \left( \hat{\sigma}_j(n) \right) \right),
$$

where $\hat{\pi}_j^*(n) = 2\Phi(-|T_j^*(n)|)$ and $T_j^*(n) = \sqrt{n}(\hat{X}_j(n) - \mu_{0,j(n)})/\sigma_j(n)$.

Now the integration by parts formula (24) combined with theorem 1 yields

$$
\left\| \max_{1 \leq j \leq p_n} \left| \frac{\sqrt{n}(\hat{X}_j(n) - \mu_j(n))}{\sigma_j(n)} + \frac{\int_{[a_j(n), b_j(n)]} B_j(n)(F_j(n)(x))dx}{\sigma_j(n)} \right| \right\|_{\psi_1}
\leq \left( \frac{c_1 \log n + c_2 \log p_n}{\sqrt{n}} \right) \max_{1 \leq j \leq p_n} \left| \frac{b_j(n) - a_j(n)}{\sigma_j(n)} \right|,
$$

where $c_1, c_2$ and $B_1(n), \ldots, B_{p_n(n)}$ are as given in theorem 2 and where

$$
Z_{j(n)} = -\int_{[a_j(n), b_j(n)]} B_j(n)(F_j(n)(x))dx/\sigma_j(n)
$$

is standard normal for all $1 \leq j \leq p_n$. This, combined with the fact that $|\Phi(x) - \Phi(y)| \leq |x - y|/2$ for all $x, y \in \mathbb{R}$, yields the desired result. □

**Proof of corollary 3** That the left-hand-side of (2) is $o_P(1)$ follows from condition 5 combined with theorem 1. By the definition of the sample median, we have that $\hat{F}_j(n)(\hat{\xi}_j(n)) - F_j(n)(\xi_j(n)) = E_j(n)$, where $|E_j(n)| \leq 1/n$. This now implies that $\hat{F}_j(n)(\hat{\xi}_j(n)) - F(\hat{\xi}_j(n)) + F(\hat{\xi}_j(n)) = E_j(n)$. The result now follows from the mean value theorem and condition 8. □

**Proof of Corollary 4** Now, for some $\xi_j^*(n)$ in between $\xi_j(n)$ and $\hat{\xi}_j(n)$, we have $f_j(n)(\xi_j^*(n))(\hat{\xi}_j(n) - \xi_j(n)) = -f_j(n)(\hat{\xi}_j(n)) + f_j(n)(\xi_j(n))$. Using the conditions of the corollary, we obtain that the $f_j(n)$ terms are simultaneously consistent for the quantities $f_j(n)(\xi_j^*(n))$ and that these later quantities are bounded above and below. Now we can argue as in the first part of the proof of corollary 2 that $\max_{1 \leq j \leq p_n} |\hat{\pi}_j'(n)| -
\[ \hat{\pi}'_{j(n)} = \alpha_P(1), \text{ where } \hat{\pi}'_{j(n)} = 2\Phi(-|\hat{U}_{j(n)}|) \text{ and} \]

\[ \hat{U}_{j(n)} = 2\sqrt{n} f_j(n)(\xi'_j(n) - \xi_{0,j(n)}) \]
\[ = -2\sqrt{n}(F_j(n)(\hat{\xi}_j(n)) - F_j(n)(\xi_j(n))) + 2\sqrt{n}f_j(n)(\xi'_j(n))(\xi_j(n) - \xi_{0,j(n)}). \]

Note that

\[ \sqrt{n} \left( F_j(n)(\hat{\xi}_j(n)) - F_j(n)(\xi_j(n)) \right) \]
\[ = -\sqrt{n} \left( \hat{F}_j(n)(\hat{\xi}_j(n)) - \hat{F}_j(n)(\xi_j(n)) - \hat{F}_j(n)(\xi_j(n)) + F_j(n)(\xi_j(n)) \right) \]
\[ - \sqrt{n} \left( \hat{F}_j(n)(\xi_j(n)) - F_j(n)(\xi_j(n)) \right) + \sqrt{n} \left( \hat{F}_j(n)(\xi_j(n)) - F_j(n)(\xi_j(n)) \right) \]
\[ \equiv -A_j(n) - V_j(n) + C_j(n), \]

where \( C_j(n) = \sqrt{n}E_j(n) \) and \( E_j(n) \) as defined in the proof of corollary \( \Box \) with \( |E_j(n)| \leq 1/n \). Hence \( C_j(n) \) vanishes asymptotically, uniformly over \( 1 \leq j \leq p_n \). Theorem \( \Box \) tells us that we can, uniformly over \( 1 \leq j \leq p_n \), replace \( A_j(n) \) and \( V_j(n) \) with \( A'_j(n) = B_j(n)(F_j(n)(\hat{\xi}_j(n))) - B_j(n)(F_j(n)(\xi_j(n))) \) and \( V'_j(n) = B_j(n)(1/2) \). Note that \( Z_j(n) \equiv 2B_j(n)(1/2) \) are standard normals and that \( B_j(n)(t) = W_j(n)(t) - tW_j(n)(1) \), for all \( t \in [0,1] \), for some standard Brownian motions \( W_j(n) \). Thus, by the symmetry properties of Brownian motion, \( |A'_j(n)| \)

\[ \leq \sqrt{\delta_{j(n)}} \left[ \sup_{0 \leq t \leq \delta_{j(n)}} |W_j(n)(t)| + \sup_{0 \leq t \leq \delta_{j(n)}} |W''_j(n)(t)| \right] + \delta_{j(n)}|W_j(n)(1)| \equiv \tilde{A}_{j(n)}(\tilde{\delta}_{j(n)}), \]

where \( \tilde{\delta}_{j(n)} \equiv M|\hat{\xi}_j(n) - \xi_j(n)|; \) \( M \) is as defined in \( \Box \); and where \( W_j(n), W'_j(n) \) and \( W''_j(n) \) are Brownian motions.

Now, for each \( k < \infty \) and \( \rho > 0 \), we have

\[ P \left( \max_{1 \leq j \leq p_n} |A'_j(n)| > \rho \right) \]
\[ \leq P \left( \max_{1 \leq j \leq p_n} \tilde{A}_{j(n)}(k\tau_{n}) > \rho \right) + P \left( \max_{1 \leq j \leq p_n} \tilde{\delta}_{j(n)} > k\tau_{n} \right), \]
where \( r_n \equiv \log(n \lor p_n)/n + \sqrt{\log p_n}/n \). However, using the facts that a standard normal deviate and the supremum of the absolute value of a Brownian motion over \([0,1]\) both have sub-Gaussian tails (i.e., have bounded \( \psi_2 \)-norms), we have \( \max_{1 \leq j \leq p_n} A_j(n)(kr_n) \leq O_p \left( \sqrt{\log p_n} \left[ r_n + \sqrt{r_n} \right] \right) \to 0 \), in probability, since \( \log^3 p_n/n \to 0 \). Thus the first term on the right-hand-side of (26) also goes to zero as \( n \to \infty \). Since corollary 3 implies \( \lim_{k \to \infty} \max_{1 \leq j \leq p_n} \delta_j(n) > kr_n \) goes to zero. Since corollary 3 implies \( \lim_{k \to \infty} \max_{1 \leq j \leq p_n} \delta_j(n) > kr_n \) goes to zero, the left-hand-side of (26) also goes to zero as \( n \to \infty \). Thus \( \tilde{U}_j(n) \) can be approximated by \( U'_j(n) = Z_j(n) + 2\sqrt{n}f_j(n)(\xi_j^*(n))(\xi_j(n) - \xi_{0,j}(n)) \) simultaneously over all \( 1 \leq j \leq p_n \).

Now we can use arguments given at the beginning of the proof of corollary 2 (again) in combination with the simultaneous consistency of \( \hat{\xi}_j(n) \) and the assumed properties of \( f_j(n) \) to obtain that

\[
\max_{1 \leq j \leq p_n} \left| \Phi(-|U'_j(n)|) - \Phi \left( -\frac{f_j(n)(\xi_j(n))}{f_j(n)(\xi_j^*(n))}Z_j(n) + 2\sqrt{n}f_j(n)(\xi_j(n))(\xi_j(n) - \xi_{0,j}(n)) \right) \right| = o_P(1).
\]

By condition (11), we have that

\[
\max_{1 \leq j \leq p_n} \left| \frac{f_j(n)(\xi_j(n))}{f_j(n)(\xi_j^*(n))} - 1 \right| \leq O_p \left( \max_{1 \leq j \leq p_n} |Z_j(n)| \right) \times \max_{1 \leq j \leq p_n} \sup_{w \leq \eta_n, |u| \leq \epsilon} \left| \frac{f_j(n)(\xi_j(n) + u) - f_j(n)(\xi_j(n))}{\epsilon^{1/2}} \right| \eta_n^{1/2} \leq O_p(\sqrt{\log p_n}) \times \eta_n^{1/2} = O_p \left( \sqrt{\log p_n} \times \sqrt{\frac{\log n \lor p_n}{n} + \sqrt{\frac{\log p_n}{n}}} \right) \to 0,
\]

where the equality follows from corollary 3. The desired result now follows. □

**Proof of corollary 5.** The proof follows the same general logic as the proof of corollary 2. Using the fact that, for any \( x \in \mathbb{R} \) and any \( y > 0 \), \( |\Phi(xy) - \Phi(x)| \leq \)
0.25 \times |1 - y| \lor |1 - y^{-1}|$, we have

$$
(27) \max_{1 \leq j \leq p_n} \left| \hat{\pi}^*_j(n) - \tilde{\pi}^{**}_j(n) \right| \leq \max_{1 \leq j \leq p_n} \frac{1}{2} \left\{ \left( \frac{n_1 \left[ \frac{(2)}{\sigma_j(n)} \right]^2 + n_2 \left[ \frac{(1)}{\sigma_j(n)} \right]^2}{n_1 \left[ \frac{(2)}{\sigma_j(n)} \right]^2 + n_2 \left[ \frac{(1)}{\sigma_j(n)} \right]^2} \right)^{1/2} - 1 \right\} \lor \left\{ \left( \frac{n_1 \left[ \frac{(2)}{\sigma_j(n)} \right]^2 + n_2 \left[ \frac{(1)}{\sigma_j(n)} \right]^2}{n_1 \left[ \frac{(2)}{\sigma_j(n)} \right]^2 + n_2 \left[ \frac{(1)}{\sigma_j(n)} \right]^2} \right)^{1/2} - 1 \right\},
$$

where $\hat{\pi}^{**}_j(n) = 2\Phi(-|T^{**}_{j(n)}|)$ and

$$
T^{**}_{j(n)} = \left( \frac{n_2 \left[ \frac{(1)}{\sigma_j(n)} \right]^2}{n_1 \left[ \frac{(2)}{\sigma_j(n)} \right]^2 + n_2 \left[ \frac{(1)}{\sigma_j(n)} \right]^2} \right)^{1/2} \sqrt{n_1} \left( \frac{\hat{X}^{(1)}_{j(n)} - \mu^{(1)}_{j(n)}}{\sigma^{(1)}_{j(n)}} \right) - \left( \frac{n_1 \left[ \frac{(2)}{\sigma_j(n)} \right]^2}{n_1 \left[ \frac{(2)}{\sigma_j(n)} \right]^2 + n_2 \left[ \frac{(1)}{\sigma_j(n)} \right]^2} \right)^{1/2} \sqrt{n_2} \left( \frac{\hat{X}^{(2)}_{j(n)} - \mu^{(2)}_{j(n)}}{\sigma^{(2)}_{j(n)}} \right) + \sqrt{\frac{n_1n_2}{n_1 \left[ \frac{(2)}{\sigma_j(n)} \right]^2 + n_2 \left[ \frac{(1)}{\sigma_j(n)} \right]^2}} \left( \mu^{(1)}_{j(n)} - \mu^{(2)}_{j(n)} \right).
$$

Now, virtually identical Brownian bridge approximation arguments to those used in the proof of corollary 2 yield that

$$
\max_{1 \leq j \leq p_n} \left| \hat{\pi}^{**}_j(n) - \pi^*_j(n) \right| \leq \sum_{k=1,2} c_1 \log n_k + c_2 \log p_n \left( \max_{1 \leq j \leq p_n} \frac{|\hat{b}^{(k)}_{j(n)} - a^{(k)}_{j(n)}|}{\sigma^{(k)}_{j(n)}} \right).
$$

In order to finish the proof, we need to bound the right-hand-side of (27). To
begin with, note that for any scalars $c_1, c_2, d_1, d_2 \geq 0$,

\[
\left| \frac{n_1 c_2^2 + n_2 c_1^2}{n_1 d_2^2 + n_2 d_1^2} \right|^{1/2} - 1 \leq \left| \left( \frac{n_1 c_2^2 + n_2 c_1^2}{n_1 d_2^2 + n_2 d_1^2} \right)^{1/2} - \left( \frac{n_1 c_2^2 + n_2 d_1^2}{n_1 d_2^2 + n_2 d_1^2} \right)^{1/2} \right| \\
+ \left| \left( \frac{n_1 c_2^2 + n_2 d_1^2}{n_1 d_2^2 + n_2 d_1^2} \right)^{1/2} - \left( \frac{n_1 d_2^2 + n_2 d_1^2}{n_1 d_2^2 + n_2 d_1^2} \right)^{1/2} \right| \\
\leq \left( \frac{n_2 d_1^2}{n_1 d_2^2 + n_2 d_1^2} \right)^{1/2} \left| \frac{c_1}{d_1} - 1 \right| \\
+ \left( \frac{n_1 d_2^2}{n_1 d_2^2 + n_2 d_1^2} \right)^{1/2} \left| \frac{c_2}{d_2} - 1 \right| \\
\leq \left| \frac{c_1}{d_1} - 1 \right| + \left| \frac{c_2}{d_2} - 1 \right| ,
\]

where the second inequality follows from the fact that for any $a, b, x, y \geq 0$,

\[
\left| (ax^2 + b)^{1/2} - (ay^2 + b)^{1/2} \right| \leq \sqrt{a} |x - y| .
\]

Hence both

\[
\left| \left( \frac{n_1 \sigma_{j(n)}^{(2)}}{\sigma_{j(n)}^{(1)}} \right)^2 + n_2 \left( \frac{n_1 \sigma_{j(n)}^{(2)}}{\sigma_{j(n)}^{(1)}} \right)^2 \right|^{1/2} - 1 \leq \left| \frac{\sigma_{j(n)}^{(1)}}{\sigma_{j(n)}^{(1)}} - 1 \right| + \left| \frac{\sigma_{j(n)}^{(2)}}{\sigma_{j(n)}^{(1)}} - 1 \right|
\]

and

\[
\left| \left( \frac{n_1 \sigma_{j(n)}^{(2)}}{\sigma_{j(n)}^{(1)}} \right)^2 + n_2 \left( \frac{n_1 \sigma_{j(n)}^{(2)}}{\sigma_{j(n)}^{(1)}} \right)^2 \right|^{1/2} - 1 \leq \left| \frac{\sigma_{j(n)}^{(1)}}{\sigma_{j(n)}^{(1)}} - 1 \right| + \left| \frac{\sigma_{j(n)}^{(2)}}{\sigma_{j(n)}^{(1)}} - 1 \right| ,
\]

and thus the right-hand-side of (27) is bounded by

\[
\frac{1}{2} \sum_{k=1,2} \max_{1 \leq j \leq p_n} \left( \frac{\sigma_{j(n)}^{(k)}}{\sigma_{j(n)}^{(1)}} \lor \frac{\sigma_{j(n)}^{(k)}}{\sigma_{j(n)}^{(2)}} \right) \left| \frac{1}{\sigma_{j(n)}^{(k)}} - \frac{1}{\sigma_{j(n)}^{(k)}} \right| ,
\]

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completing the proof. □

*Proof of corollary 6.* The proof consists of extending the proof of corollary 4 in a manner similar to the way in which the proof of corollary 2 was extended for proving corollary 5. A key difference is that the role of $\sigma_j^{(k)}(n)$ and $\hat{\sigma}_j^{(k)}(n)$ in the proof of corollary 5 is replaced by $1/f_j^{(k)}(n)$ and $1/\hat{f}_j^{(k)}(n)$, for $k = 1, 2$ and $1 \leq j \leq p_n$. The remaining necessary extensions of the proof of corollary 4 are straightforward. □

*Proof of lemma 1.* Because of the invariance under monotone transformation, we can assume without loss of generality that the data are uniformly distributed. Classical arguments in Billingsley (1968) yield the second result. In particular, the form of the limiting distribution function, which is the distribution of the supremum in absolute value of a Brownian bridge, can be found on page 85 of Billingsley. Arguments for establishing the remaining two results can be found in section 3.9.4 (for the Wilcoxon statistic) and in section 2.13.2 (for the Cramér-von Mises statistic) of van der Vaart and Wellner (1996). □

*Proof of theorem 3.* Define $\tilde{E}_n = \max_{1 \leq j \leq p_n} \|\tilde{F}_j(n) - \hat{F}_j(n)\|_\infty$ and, for each $\delta \geq 0$, $\tilde{E}_n(\delta) = \max_{1 \leq j \leq p_n} \sup_{|s-t| \leq \delta} \left|\tilde{F}_j(n)(s) - \hat{F}_j(n)(t)\right|$. Suppose now that for some positive, non-increasing sequences $\{s_n, \delta_n\}$, with $\delta_n \to 0$, we have $\tilde{E}_n(\delta_n) = o_p(s_n)$ and $P(\tilde{\epsilon}_n > \delta_n) = o(1)$. Then, by the definition of $\tilde{\epsilon}_n$,

$$
\tilde{E}_n(n) = \tilde{E}_n 1\{\tilde{\epsilon}_n \leq \delta_n\} + \tilde{E}_n 1\{\tilde{\epsilon}_n > \delta_n\} \leq \tilde{E}_n(\delta_n) + o_p(s_n) = o_p(s_n).
$$

Now, by theorem 2 and condition (21), we have for any sequence $\delta_n \downarrow 0$,

$$
\sqrt{n}E_n(\delta_n) \leq \max_{1 \leq j \leq p_n} \sup_{|s-t| \leq \delta_n} \sqrt{n} \left|\tilde{F}_j(n)(s) - F_j(n)(s) - \hat{F}_j(n)(t) + F_j(n)(t)\right| + \sqrt{n} M \delta_n
$$

$$
\leq \max_{1 \leq j \leq p_n} \sup_{|s-t| \leq \delta_n} \left|B_j(n)(F_j(n)(s)) - B_j(n)(F_j(n)(t))\right|
$$

$$
+ O_P \left(\frac{\log n + \log p_n}{\sqrt{n}} + \sqrt{n} \delta_n\right).
$$

Combining this with a reapplication of condition (21) along with lemma 2 below (a
precise modulus of continuity bound for Brownian motion), we obtain

\[
\sqrt{n} \hat{E}_n(\delta_n) \leq O_P \left( \sqrt{\log p_n \delta_n \log(1/\delta_n)} + \frac{\log n + \log p_n}{\sqrt{n}} + \sqrt{n} \delta_n \right).
\]

(29)

Both (28) and (29) will prove useful at several points in our proof.

Using the fact that \( \hat{\epsilon}_n = o_P(1) \), we can find a positive, sufficiently slowly decreasing sequence \( \delta_n \rightarrow 0 \) such that \( \hat{\epsilon}_n = o_P(\delta_n) \). Now, by applying (28) with \( s_n = 1 \), we obtain result (i) of the theorem: \( \hat{E}_n = o_P(1) \). For result (ii), we can use the fact that \( \log \frac{2}{p_n}/n = o(1) \), to construct a positive, non-decreasing sequence \( r_n \rightarrow \infty \) slowly enough so that \( r_n \log p_n/\sqrt{n} = o(1) \) and \( r_n/\log n = o(1) \). Since \( \sqrt{n}(\log n)\hat{\epsilon}_n = O_P(1) \), we have

\[
\sqrt{n} \hat{\epsilon}_n \log(1/\hat{\epsilon}_n) = \sqrt{n}(\log n)\hat{\epsilon}_n \left( \frac{\log \sqrt{n} - \log(\sqrt{n}\hat{\epsilon}_n)}{\log n} \right) = O_P(1).
\]

Thus, if we set \( \delta_n = r_n/(\sqrt{n}\log n) \), we have \( \hat{\epsilon}_n = o_P(\delta_n) \). We also have, by (29), that

\[
\hat{E}_n(\delta_n) = O_P \left( \sqrt{\frac{1}{n} \times \frac{r_n \log p_n}{\sqrt{n}} \times \frac{\log \sqrt{n} + \log \log n - \log r_n}{\log n}} + o \left( \frac{1}{\sqrt{n}} \right) \right) = o_P(n^{-1/2}).
\]

The proof is now complete by reapplying (28) with the choice \( s_n = n^{-1/2} \).

**Lemma 2** Let \( W : [0, 1] \mapsto \mathbb{R} \) be a standard Brownian motion. Then there exists a universal constant \( k_0 < \infty \) such that

\[
\left\| \sup_{|s-t| \leq \delta} |W(s) - W(t)| \right\|_{\psi_2} \leq k_0 \sqrt{\delta \log(1/\delta)}
\]

for all \( 0 < \delta \leq 1/2 \).

**Proof.** Fix \( \delta \in (0, 1) \). Let \( n_\delta \) be the smallest integer \( \geq 1 + 1/\delta \), and extend the
Brownian motion $W$ to the interval $[0, \delta n]$. Now

\begin{equation}
\sup_{|s-t| \leq \delta} |W(s) - W(t)| \leq \max_{1 \leq j \leq n} \sup_{(j-1)\delta \leq s \leq t \leq (j+1)\delta} |W(s) - W(t)|
\leq 2 \max_{1 \leq j \leq n} \sup_{t \in [(j-1)\delta, (j+1)\delta]} |W(t) - W((j-1)\delta)|
\leq 2 \max_{1 \leq j \leq n} \sup_{1 \leq n} \sqrt{2\delta} |W_j^*(t)|,
\end{equation}

where $W_1^*, \ldots, W_n^*$ are a dependent collection of standard Brownian motions. The last inequality follows from the symmetry properties of Brownian motion. We can now use the fact that the tail probabilities of the supremum over $[0, 1]$ of the absolute value of Brownian motion are sub-Gaussian (and thus have bounded $\psi_2$-norms) to obtain that the $\psi_2$-norm of the left side of (30) is bounded by $k^* \sqrt{2} \delta \log(1 + 1/\delta)$, where $k^* = 5k_*$ does not depend on $\delta$. The last inequality follows because $\log(3 + 1/\delta)/\log(1/\delta) \leq 1 + \log(1 + 3/\delta)/\log(1/\delta) \leq 3$ for all $\delta \in (0, 1/2]$. \hfill \Box

Proof of corollary 7. Result (i) follows directly from part (i) of theorem 3 and theorem 1. Result (ii) is a direct consequence of part (i) of theorem 3 and a minor modification of the integration by parts identity (24) used in the proof of corollary 2. The proof of result (iii) is a straightforward extension of the proof of corollary 3 which incorporates the conclusion of part (i) of theorem 3. \hfill \Box

Proof of corollary 8. For result (i), we use part (ii) of theorem 3 combined with integration by parts to obtain that

$$\max_{1 \leq j \leq p_n} |\bar{X}_j(n) - \bar{X}_j(n)| = o_P \left( n^{-1/2} \left[ \max_{1 \leq j \leq p_n} |b_j(n) - a_j(n)| + 2\epsilon_n \right] \right) = o_P(1).$$

Now corollary 1 gives us the desired results since

$$\sqrt{\frac{\log p_n}{n}} \max_{1 \leq j \leq p_n} |b_j(n) - a_j(n)| = \sqrt{\frac{\log p_n}{n}} \max_{1 \leq j \leq p_n} n^{1/4} |b_j(n) - a_j(n)| = o(1).$$

For result (ii), we also use part (ii) of theorem 3 combined with integration by parts...
to obtain

\[
\max_{1 \leq j \leq p_n} \left| \frac{\sqrt{n} (\bar{X}_{j(n)} - \bar{X}_{j(n)})}{\sigma_{j(n)}} \right| \leq o_P \left( \max_{1 \leq j \leq p_n} \frac{|b_{j(n)} - a_{j(n)}| + 2\epsilon_n}{\sigma_{j(n)}} \right) = o_P(1),
\]

and the desired result follows using the Brownian bridge approximation of \( \sqrt{n} \left( \bar{X}_{j(n)} - \mu_{j(n)} \right) / \sigma_{j(n)} \) given in the proof of corollary \( \mathbb{2} \). For result (iii), the desired conclusion is obtained via part (ii) of theorem \( \mathbb{3} \) combined with a straightforward adaptation of the proof of corollary \( \mathbb{4} \).

**Proof of corollary \( \mathbb{4} \).** The proof follows almost immediately from applying part (ii) of theorem \( \mathbb{3} \) to each sample separately, yielding the result

\[
\max_{k=1,2} \max_{1 \leq j \leq p_n} \sqrt{n_k} \left\| \hat{F}_{j(n)}^{(k)} - \hat{F}_{j(n)}^{(k)} \right\|_\infty = o_P(1).
\]

Now, the proofs of results (i) and (ii) are direct extensions of the one-sample results of corollary \( \mathbb{3} \) combined with straightforward adaptations of arguments found in the proofs of corollaries \( \mathbb{5} \) and \( \mathbb{6} \). The proof of result (iii) also follows almost immediately. For the Kolmogorov-Smirnov statistic, the result is obvious. For the other two statistics, the result follows with some help from integration by parts. □

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Table 1. One sample simulation study results for the mean, median and signed rank statistics under models 1, 2 and 3. Tot.: total count identified using FDR. Pos.: number of true positives identified using FDR. EFDR: empirical FDR.

| Model | Mean | Median | Signed rank |
|-------|------|--------|-------------|
|       | Tot.(Pos.) | EFDR | Tot.(Pos.) | EFDR | Tot.(Pos.) | EFDR |
|-------|-------------|------|-------------|------|-------------|------|
|       | Sample size = 20 | | | | |
| 1     | 64.7(33.9)  | 0.47 | 31.8(25.4)  | 0.19 | 15.5(15.5)  | 0.01 |
| 2     | 64.4(33.9)  | 0.47 | 31.6(25.3)  | 0.19 | 15.3(15.2)  | 0.01 |
| 3     | 64.0(33.9)  | 0.46 | 31.1(25.0)  | 0.19 | 15.2(15.1)  | 0.01 |
|-------|-------------|------|-------------|------|-------------|------|
|       | Sample size = 50 | | | | |
| 1     | 54.2(37.8)  | 0.30 | 38.7(32.9)  | 0.15 | 34.5(34.0)  | 0.01 |
| 2     | 53.7(37.4)  | 0.29 | 38.5(32.7)  | 0.14 | 34.2(33.8)  | 0.01 |
| 3     | 52.3(37.5)  | 0.27 | 38.2(32.5)  | 0.14 | 34.4(33.9)  | 0.01 |
Table 2. Two sample simulation study results for mean, median, Wilcoxon and Kolmogorov-Smirnov (KS) statistics under model 4. Tot.: total count identified using FDR. Pos: number of true positives identified using FDR. EFDR: empirical FDR.

| Mean | Median | Wilcoxon | KS |
|------|--------|----------|----|
| Tot.(Pos.) | EFDR | Tot.(Pos.) | EFDR | Tot.(Pos.) | EFDR | Tot.(Pos.) | EFDR |
| $n_1 = n_2 = 10$ | $47.3(21.5)$ | 0.54 | $8.4(6.7)$ | 0.18 | $14.4(13.1)$ | 0.08 | $2.6(2.4)$ | 0.08 |
| $n_1 = n_2 = 30$ | $40.9(28.9)$ | 0.28 | $21.2(19.8)$ | 0.06 | $32.0(26.6)$ | 0.16 | $23.7(21.5)$ | 0.09 |
| $n_1 = n_2 = 60$ | $43.4(33.3)$ | 0.22 | $29.7(25.4)$ | 0.14 | $39.4(32.4)$ | 0.17 | $32.1(28.0)$ | 0.12 |
Figure 1: Estrogen data. Scatter plots of p-values comparing the four approaches (mean, median, Wilcoxon and KS). A lowess smoother is used to estimate the trend, and the associated rank correlation coefficient (tau) is given above each panel.