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A more flexible parametric estimation of univariate reference intervals: A new method based on the GS-distribution

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Abstract

Background: Reference interval estimation is an important issue in clinical laboratories. Present methods are based either on data transformation or on non-parametric approaches.
Methods: We present a new technique based in a family of statistical distributions known as GS-distributions that provide a suitable model for continuous unimodal variables. We compare, both by simulation studies an on actual data, the reference intervals estimated by using non-parametric methods and data transformations suggested by the IFCC and those obtained by fitting a GS-distribution. Simulated data are generated from various distributions to evaluate the accuracy of these methods. In each case, confidence intervals for the resulting reference intervals are obtained by bootstrap.
Results: In all the cases, the GS-distribution based method provides comparable or more accurate results than the non-parametric methods. In most cases, the proposed method produces better results than those obtained by transforming the original data.
Conclusions: Our results suggest that the method for computing reference intervals based on GS-distribution is a valid alternative for the current non-parametric methods.

Keywords: Reference intervals; Statitical distributions; Reference range

1. Introduction

Reference intervals (RI) play an important role in clinical practice as they are required for assessing the health status of patients. Furthermore, they are a basic tool of clinical laboratories, both in quality control and in providing reference values according to the protocols used in each case. An RI is typically defined as the range comprised between the 2.5 and 97.5 percentiles of the data distribution from a given reference population. Accordingly this interval estimates the expected values that would contain the 95% of the subjects of the considered population. Guidelines for appropriately estimating RI include rules for subject selection, data validation, outlier detection, and indications on the appropriate statistical computations [1]. In particular, the target reference population should be clearly defined. In general, the reference population is any population defined according to precise inclusion criteria and it does not always correspond strictly to a healthy population [2]. For instance, in a given clinical application the target population could be those patients of a given age range that present a severe status of a given disease.

From a statistical point of view, the available approaches for RI estimation include non-parametric methods [3–5], robust methods [6–8], transformation methods [9–11], and different variants of these basic methodologies (see Ref. [12] for a review). From a practical point of view, it is common to follow the NCCLS (National Committee for Clinical Laboratory Standards) recommendations and obtain nonparametric reference intervals using a sample size of at least 120 subjects [1].

The development of the different methods indicated above arises from the lack of information on the underlying

Abbreviations: GSD, GS-distribution; TST, Two-stages transformation method; NP, non-parametric method; HDM, Harrell–Davis method; RMSE, Root Mean Square Error.

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distribution. Otherwise, the computation of the corresponding RI would be straightforward once the appropriated parameters for the distribution were obtained from the sample data. When no reasonable information is available on the underlying distribution, one may consider using a transformation that provides a new variable with know distribution. For instance, an appropriate Box–Cox transformation can convert the actual variable in a new variable with normal distribution [10,11]. RI would then be estimated on the transformed variable; a reverse transformation would provide the required RI on the original variable. In many cases, a simple logarithmic transformation is used [13,14]. However, this may not be appropriate for most situations and alternative transformations should be considered. Although this is a suitable technique, there is no guarantee that this transformation exists for a given set of data [15]. Furthermore, even when the appropriate transformation can be found, the back transformation may be impossible, due to out-of-bounds problems with interpolated values. Non-parametric methods provide a highly recommended alternative in those cases.

The problem of estimating a RI would be greatly simplified if a general parametric model could be defined. Then, the problem would be reduced to obtain the appropriate parameters according to the data and to assess the goodness-of-fit. Once the particular instance of the distribution is fitted, RI would be obtained by a simple computation. With this possibility in mind, we developed the GS-distribution (GSD) [16]. This is a family of distributions defined as:

$$\frac{\mathrm{d}F(x)}{\mathrm{d}x} = \alpha F(x)^g (1-F(x)^k)^\gamma \qquad F(x_0) = 0.5 \tag{1}$$

where $F(x)$ is the cumulative. This family has three parameters that account for the shape of the resulting distribution (g, k, γ). Thus, these parameters are responsible for the skewness and kurtosis of the resulting distribution. Parameter α is related to the spread of the distribution, and x_0 corresponds to the median of the distribution and fixes the initial conditions of the differential equation. For simplicity, we shall indicate a given GSD as GSD[x_0, α, g, k, γ]. Some examples of the flexibility of the GSD are shown in Fig. 1.

The GSD is a parametric family that results from a generalization of the S-distribution [17–19] and it is more flexible than classical parametric models and hence is better for modeling data observed in practice [16]. Using this family, we can fit a GSD to unimodal data without further assumptions on the actual underlying distribution. As any continuous unimodal distribution can be accurately represented as a GSD, this assures that we can always obtain an estimated distribution that fit the data. In most cases, the resulting fit is comparable to the one we would obtain if the true distribution was known. In that sense, the GSD is a practical tool for obtaining a distribution that explains the

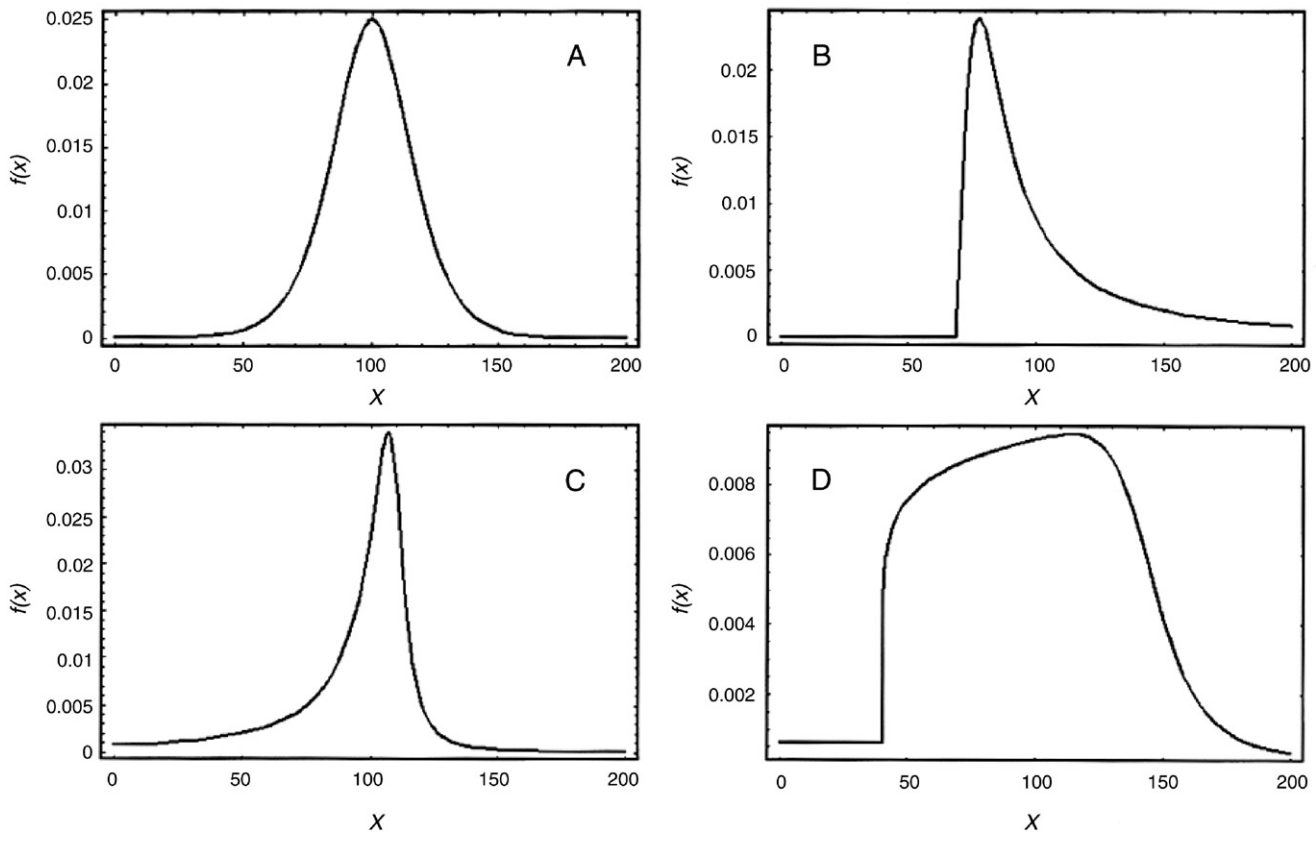

Fig. 1. Examples of GSD. In all cases the median is equal to 100. Parameters α, g, k, γ are: (A) 0.1, 1.0, 1.0, 1.0; (B) 0.1, 0.5, 1.0, 3.0; (C) 0.1, 2.0, 5.0, 2.0; (D) 0.01, 0.1, 12.0, 2.0.

observed data. In practice, GSD parameters must be estimated from data by a numerical maximum likelihood procedure [16,20].

In this paper we suggest using the GSD as a parametric model for computing RI. In the Methods section we will first discuss the use of GSD and the computation of quantiles and RI from a given instance. Then, we shall present the methods for fitting a GSD to data. In the Results section, we will investigate the performance of this method on simulated and actual data. To do that, we will compare the RI obtained by our method to those obtained by the common non-parametric and transformation methods. Our results will show that the GSD based methodology provides a practical alternative to the current methods.

2. The GS-distribution as a statistical model for univariate distributions: definition and computation of reference intervals

The GSD is defined as a five parameter family of distributions that arises from recasting known distributions using a representation of the form

$$f(x) = \frac{\mathrm{d}F}{\mathrm{d}x} = \Psi(F) \tag{2}$$

This representation has been used by different authors leading to quantile-based families of distributions [21–24]. We found that the representation in Eq. (1) is the simplest form within this class that includes different known distributions (exponential, uniform, symmetric Beta, some F distributions, logistic, generalized logistic, and a few others) as particular cases Table 1 in Muiño et al. [16] includes other distributions that are a particular case of the GSD. A detailed analysis of the parametric space defined by those distributions and the limit relationships between various distributions justifies using this generalization (see Muiño et al. [16] for details).

The GSD defined in Eq. (1) provides a flexible family of distributions for modeling continuous unimodal univariate data. The overall properties and methods corresponding to this family are discussed elsewhere [16]. Here, we shall briefly provide the main results needed for RI estimation. First, given a set of parameters, a quantile $F^{-1}(q)=x_q$ can be obtained as:

$$x_q = x_0 + \frac{B_{0.5^k,q^k}\left(\frac{1-g}{k}, 1-\gamma\right)}{\alpha k} \tag{3}$$

where $B_{z_1,z_2}(a,b)$ is the incomplete Beta function [see Ref. [16] for a justification of this expression]. Using this result, computation of a 95% RI is straightforward, since it corresponds to computing $x_{0.025}$ and $x_{0.975}$. As a matter of fact,

Table 1
Results of simulation studies comparing various approaches for estimating the 97.5th percentile

Sampling distribution	True 97.5th percentile	Sample size					
		120		180		300	
			RMSE		RMSE		RMSE
Chi-square(4)	11.1						
NP		10.7 (1.23)	1.313	11.0 (1.04)	1.043	11.0 (0.84)	0.845
TST		10.8 (0.91)	0.964	10.9 (0.85)	0.876	11.0 (0.65)	0.680
HDM		11.4 (1.19)	1.213	11.3 (0.98)	1.000	11.2 (0.78)	0.789
GSD		10.9 (0.87)	0.929	10.9 (0.85)	0.872	11.0 (0.63)	0.654
Chi-square (8)	17.5						
NP		17.4 (1.30)	1.310	17.4 (1.30)	1.310	17.4 (1.03)	1.043
TST		17.3 (1.02)	1.054	17.3 (1.02)	1.054	17.3 (0.82)	0.852
HDM		17.8 (1.22)	1.236	17.8 (1.22)	1.236	17.6 (0.97)	0.969
GSD		17.3 (1.19)	1.310	17.4 (0.99)	1.032	17.5 (0.80)	0.830
Chi-square (12)	23.3						
NP		22.7 (1.65)	1.769	23.2 (1.48)	1.483	23.3 (1.18)	1.187
TST		23.0 (1.38)	1.435	23.1 (1.16)	1.194	23.1 (0.93)	0.956
HDM		23.7 (1.64)	1.684	23.6 (1.37)	1.397	23.5 (1.10)	1.107
GSD		23.0 (1.37)	1.431	23.1 (1.16)	1.191	23.2 (0.91)	0.931
Gaussian (70, 10)	89.6						
NP		88.7 (2.13)	2.294	89.4 (1.86)	1.871	89.4 (1.63)	1.705
TST		89.0 (1.73)	1.820	89.1 (1.43)	1.505	89.2 (1.16)	1.228
HDM		89.9 (1.99)	2.020	88.9 (1.69)	1.703	89.6 (1.38)	1.388
GSD		89.0 (1.65)	1.812	89.2 (1.41)	1.471	89.3 (1.14)	1.184
pH*	7.54						
NP		7.54 (0.01)	0.019	7.54 (0.01)	0.015	7.54 (0.01)	0.012
TST		7.55 (0.01)	0.015	7.54 (0.01)	0.012	7.54 (0.01)	0.010
HDM		7.55 (0.01)	0.017	7.54 (0.01)	0.014	7.54 (0.01)	0.011
GSD		7.54 (0.02)	0.015	7.54 (0.01)	0.012	7.54 (0.01)	0.009

Entries are means of 1000 simulations together with SD (in parentheses). Data are generated from the indicated distributions. The pH* values are generated from a GSD[7.36,9.77,0.88,1.66,0.80] obtained by fitting actual data. (NP) Nonparametric approach. (TST) Two-stages transformation method. (HDM) Method of Harrel and Davis. (GSD) GS-Distribution method. (RMSE) Root mean squared error.

once a set of parameters is identified, we can obtain any RI of probability $(1-\alpha)$ by computing:

$$x_{\alpha/2} = x_0 + \frac{B_{0.5^k,(\alpha/2)^k}\left(\frac{1-g}{k}, 1-\gamma\right)}{\alpha k}$$

$$x_{(1-\alpha/2)} = x_0 + \frac{B_{0.5^k,(1-\alpha/2)^k}\left(\frac{1-g}{k}, 1-\gamma\right)}{\alpha k} \quad (4)$$

Then, from a practical point of view, the bottleneck for RI estimation using a GSD will be parameter estimation from data. This can be achieved by a maximum likelihood procedure as indicated in the appendix. Estimated GSD on different data sets are provided in Figs. 2 and 6.

2.1. Computational procedures

GSD computations require integrating Eq. (1). Furthermore, a numerical maximization procedure is needed for parameter estimation by maximum likelihood. We have defined a set of functions in *Mathematica*© (version 5.1) that implements all the required computations. Integration is obtained by using NDSolve. Numerical optimization is obtained by using FindMinimum. Simulations use the built-in procedures in *Mathematica*©. Simulated data sets from a GSD are obtained by generating a sample of random numbers from 0 to 1 and by applying the quartile equation (Eq. (2)). All figures are generated in *Mathematica*© by using the built-in graphical functions.

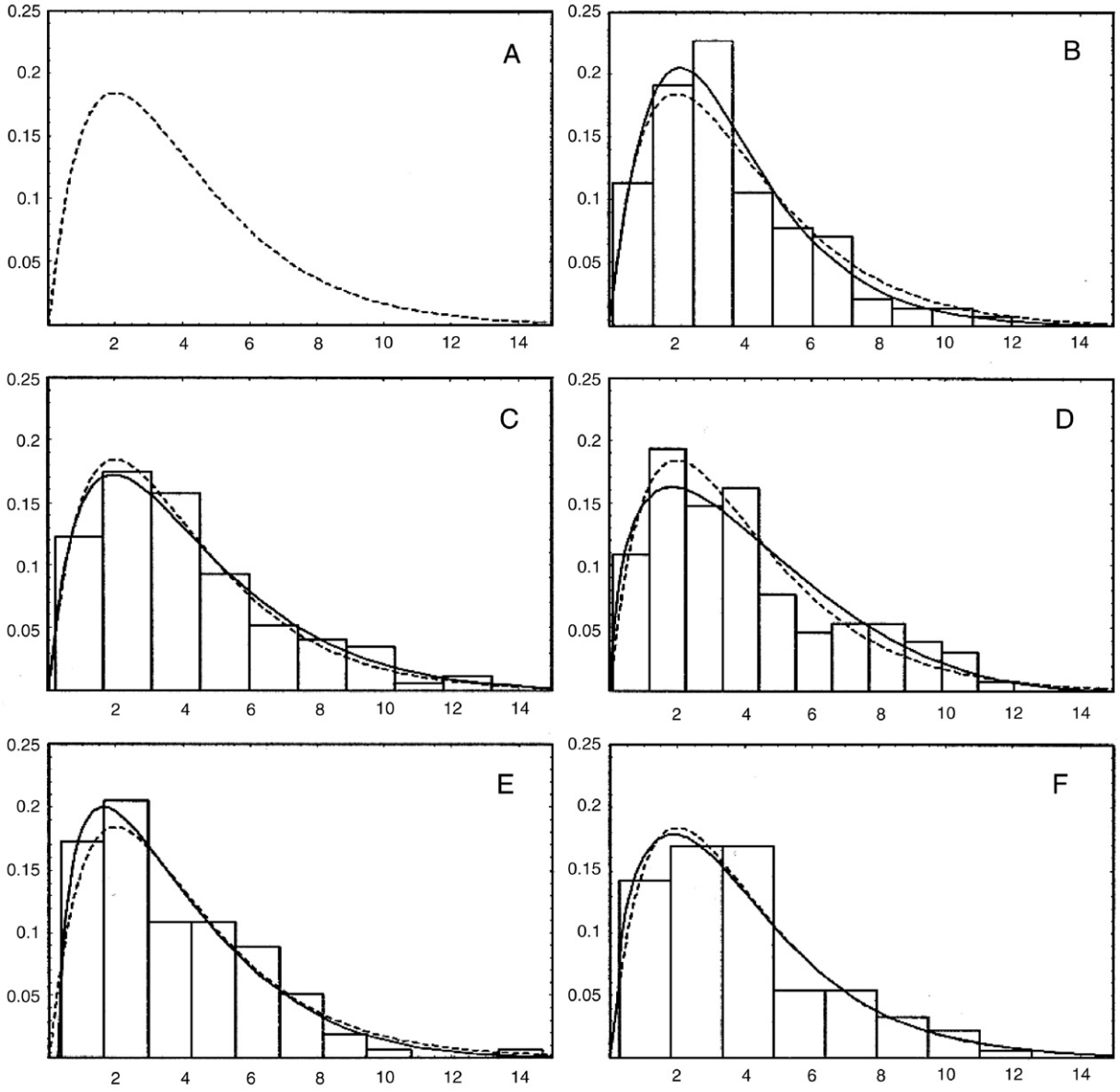

Fig. 2. Fitting a GSD to samples from a. χ^2_4 distribution (dotted line). (A) Theoretical distribution (B, C, D, E and F) histogram of the sample data (simulated from the theoretical distribution with sample size $n=120$), theoretical distribution and fitted distribution (continuous line).

3. Comparison of RI estimation methods

3.1. Simulation studies

The proposed method based on the GSD is compared with three other well-established methods for RI estimation. (i) The nonparametric (NP) approach based on the order statistics [12]; (ii) the Two-Stage Transformation (TST) recommended by the International Federation of Clinical Chemistry (IFCC) [25,26]; and (iii) the weighted percentile method of Harrell and Davis (HDM) [3]. In each case, bootstrap estimations for the RI limits are obtained. As a measure of performance for a given procedure for estimating the required percentiles we shall compute the root mean squared error (RMSE) defined as in Ref. [27]:

$$\mathrm{RMSE} = \sqrt{\frac{\sum_{i=1}^{n} (T_p - Q_p(i))^2}{n}} \tag{5}$$

where, T_p is the true p-th percentile, $Q_p(i)$ is the estimated p-th percentile in sample i, and n is the number of samples. This measure will be used for comparison purposes as it allows ranking the studied procedures [5].

As a first approach, we compared the performance of different methods of RI estimation by a simulation study (Tables 1 and 2). Random samples were generated both from Chi-square distributions, normal distribution and from GSD. The Chi-square distribution has been extensively used to model the range of unimodal positively skewed data that are generally

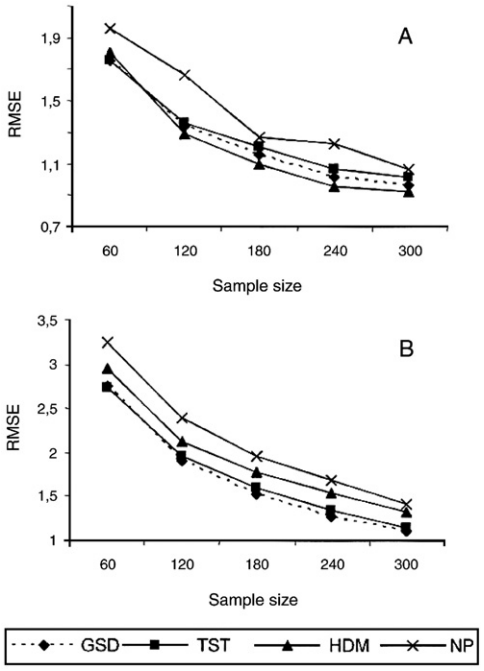

Fig. 3. Simulation study. Comparison of RI obtained by various methods: (NP) Nonparametric method. (TST) Two-stages transformation method. (HDM) Method of Harrel and Davis. (GSD) GS-Distribution method. Computations correspond to 1000 bootstrap samples for each sample size. Samples are generated from: (A) χ^2_4; (B) $N(70, 10)$.

Table 2
Results of simulation studies comparing various approaches for estimating the 2.5th percentile

Sampling distribution	True 2.5th percentile	Sample size					
		120		180		300	
			RMSE		RMSE		RMSE
Chi-square (4)	0.48						
NP		0.57 (0.14)	0.168	0.50 (0.12)	0.116	0.49 (0.09)	0.092
TST		0.54 (0.13)	0.141	0.53 (0.10)	0.117	0.51 (0.07)	0.080
HDM		0.51 (0.13)	0.131	0.49 (0.11)	0.106	0.48 (0.08)	0.083
GSD		0.52 (0.12)	0.125	0.50 (0.99)	0.106	0.49 (0.07)	0.071
Chi-square (8)	2.18						
NP		2.40 (0.39)	0.451	2.22 (0.30)	0.303	2.21 (0.23)	0.236
TST		2.35 (0.32)	0.356	2.28 (0.24)	0.258	2.26 (0.18)	0.198
HDM		2.24 (0.34)	0.348	2.19 (0.27)	0.271	2.19 (0.21)	0.212
GSD		2.31 (0.32)	0.346	2.23 (0.23)	0.242	2.21 (0.18)	0.183
Chi-square (12)	4.40						
NP		4.62 (0.54)	0.584	4.46 (0.46)	0.462	4.45 (0.36)	0.363
TST		4.57 (0.46)	0.485	4.54 (0.36)	0.386	4.52 (0.28)	0.303
HDM		4.38 (0.49)	0.499	4.40 (0.41)	0.415	4.41 (0.33)	0.325
GSD		4.51 (0.45)	0.468	4.48 (0.36)	0.368	4.45 (0.28)	0.283
Gaussian (70,10)	50.4						
NP		51.1 (2.25)	2.372	50.4 (1.97)	1.974	50.4 (1.53)	1.539
TST		50.8 (1.94)	1.999	50.7 (1.55)	1.601	50.6 (1.20)	1.237
HDM		49.9 (2.22)	2.268	50.1 (1.82)	1.837	50.2 (1.41)	1.424
GSD		50.7 (1.94)	1.983	50.6 (1.54)	1.574	50.5 (1.19)	1.203
pH*	7.11						
NP		7.12 (0.03)	0.036	7.11 (0.03)	0.031	7.11 (0.02)	0.024
TST		7.12 (0.03)	0.030	7.11 (0.02)	0.024	7.11 (0.02)	0.019
HDM		7.10 (0.03)	0.036	7.11 (0.03)	0.029	7.11 (0.02)	0.022
GSD		7.11 (0.03)	0.030	7.11 (0.02)	0.024	7.11 (0.02)	0.018

Entries are means of 1000 simulations together with SD (in parentheses). Data are generated from the indicated distributions. The pH* values are generated from a GSD[7.36,9.77,0.88,1.66,0.80] obtained by fitting actual data. (NP) Nonparametric approach. (TST) Two-stages transformation method. (HDM) Method of Harrel and Davis. (GSD) GS-Distribution method (RMSE) Root mean squared error.

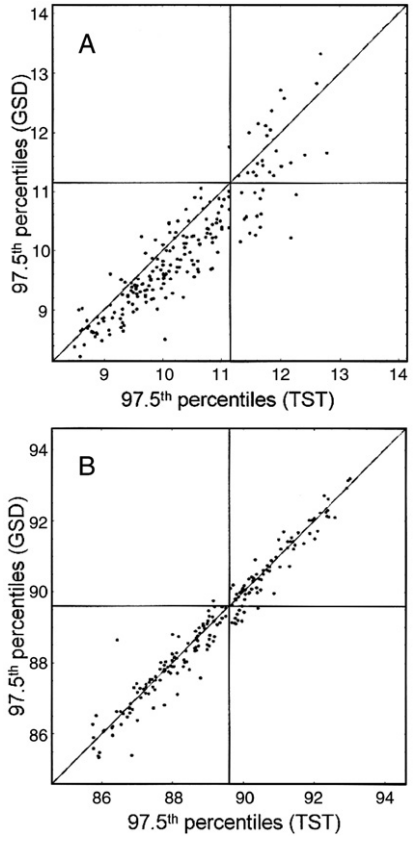

Fig. 4. Comparison of estimated percentiles: (TST) Two-stages transformation method. (GSD) GS-Distribution method. In each case, 1000 samples of size $n=120$ are generated from: χ^2_4. True percentile 97.5 (11.14). $N(70, 10)$. True percentile 97.5 (89.6).

encountered in clinical studies [4]. Following this strategy, we have used this distribution with different degrees of freedom to generate simulated data sets to test the methods for RI estimation. We have also considered data sets generated from a normal distribution and data from a GSD[7.36, 9.77, 0.88, 1.66, 0.80], that accurately represents a sample of actual pH values. In each case, 1000 samples with 120, 180, 240 and 300 values were generated.

Our results show that the GSD method provides accurate estimations with a standard deviation and an RMSE that are lower than those obtained by the other methods. As the sample size increases, the RMSE of all methods decrease (Fig. 3). The NP method is clearly worst in all conditions, while the GSD method is close to the TST results in almost all conditions. However, the GSD yields slightly better results. Only in the samples from the Chi-square distribution the HDM methods yields better results than the GSD. Results are similar by using Chi-square distributions of various degrees of freedom (not shown).

It is instructive to compare the results for the TST and GSD methods sample by sample (Fig. 4). Comparison of the estimated 97.5th percentile in samples from a Chi-square and a normal distribution shows that both methods tend to give similar results. Results are similar for the 2.5th percentile. In terms of performance, the GSD based method can be considered as equivalent to a TST. However, the GSD method provides an estimation of the distribution, which is an advantage for additional computations. Furthermore the GSD overcomes some of the potential disadvantages of the TST method, especially in those cases in which a back transformation to the original variable is not possible.

If we focus on individual samples, we observe that all methods produce similar results but with slightly different accuracies. In Fig. 5 we show the result of estimating the 90% confidence intervals for the percentiles. In each case the results correspond to 200 bootstrap samples. We choose this approach since no closer form confidence intervals for reference interval endpoints is available for all the compared methods [12]. For samples generated from a Chi-square distribution, all methods produce slightly biased estimations, with greater variances for the NP and HDM methods (Fig. 5A). For the samples generated from a normal distribution, the results are more similar between all methods (Fig. 5B).

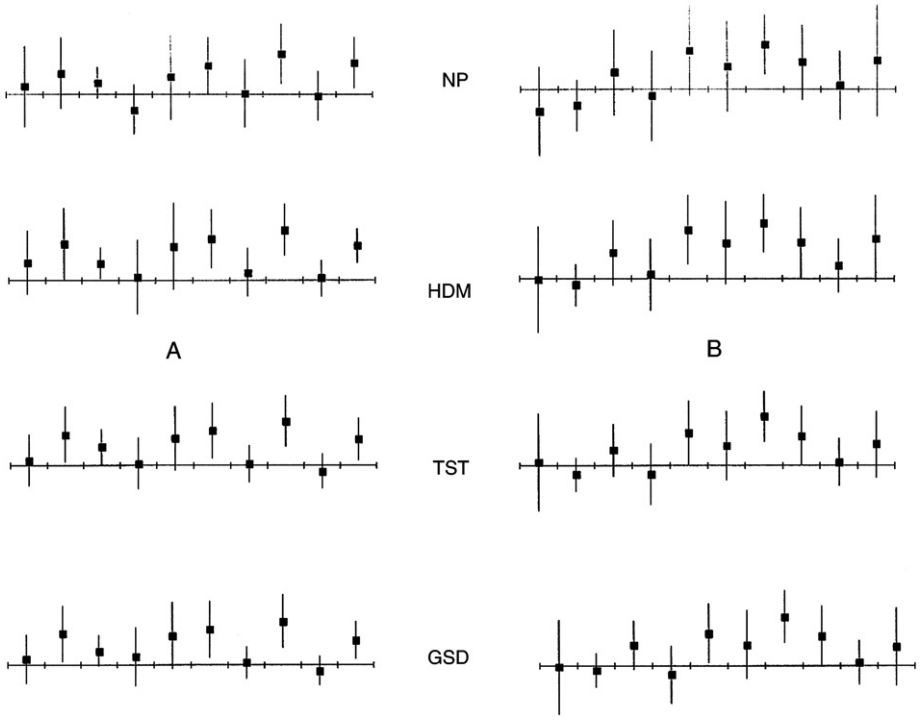

Fig. 5. Comparison of 90% confidence intervals for RI. Confidence intervals are estimated by bootstrap (200 samples for each data set). The RI for each data set is estimated by: (NP) Nonparametric method. (TST) Two-stages transformation method. (HDM) Method of Harrel and Davis. (GSD) GS-Distribution method. Data ($n=120$) are generated from: (A) χ^2_4; (B) $N(70, 10)$.

3.2. Application to clinical data

Clinical data were obtained from a data base of the Intensive Care Unit in the Hospital Universitario Arnau de Vilanova de Lleida (1997–2002). This database includes measurements on clinical and laboratory variables collected when the patient enters the Unit. The study goals include estimation of RI for the reference population of incoming patients. People older that 75 and younger than 16 were excluded. Patients with previous records on pathology were also excluded. The final sample includes 861 patients.

As an application to show the performance of the GSD method, we have selected six clinical parameters that presented different shapes. In each case, we fit a GSD as a first step before computing the RI (Fig. 6). As expected, we obtain good fits in all cases. The agreement between the fitted distribution and the sample data is shown for variables Creatinine and Leukocytes in Fig. 7. In the case of Creatinine, the TST method is not able of providing an appropriate transformation (Fig. 7B), while the GSD provides an accurate representation of the original data (Fig. 7C). Both the TST and GSD provide suitable models for the variable Leukocytes (Fig. 7E–F). From these results, one expects similar results for the RI obtained by both methods in the case of the Leukocytes, but different results for Creatinine.

RI for the selected clinical variables were obtained by the four methods (Table 3). Results are similar for all methods in the case of Urea Nitrogen, Serum Sodium and pH, with slight difference in some cases for the NP method. Again, we obtain confidence intervals for all the percentiles by bootstrapping the samples. TST and GSD produce similar results in all cases, except for Creatinine. As stated before, this can be justified because the TST does not provide a valid transformation for this variable.

4. Discussion

Reference intervals, either in the general healthy population or in a given subgroup defined by a pathology, age range,

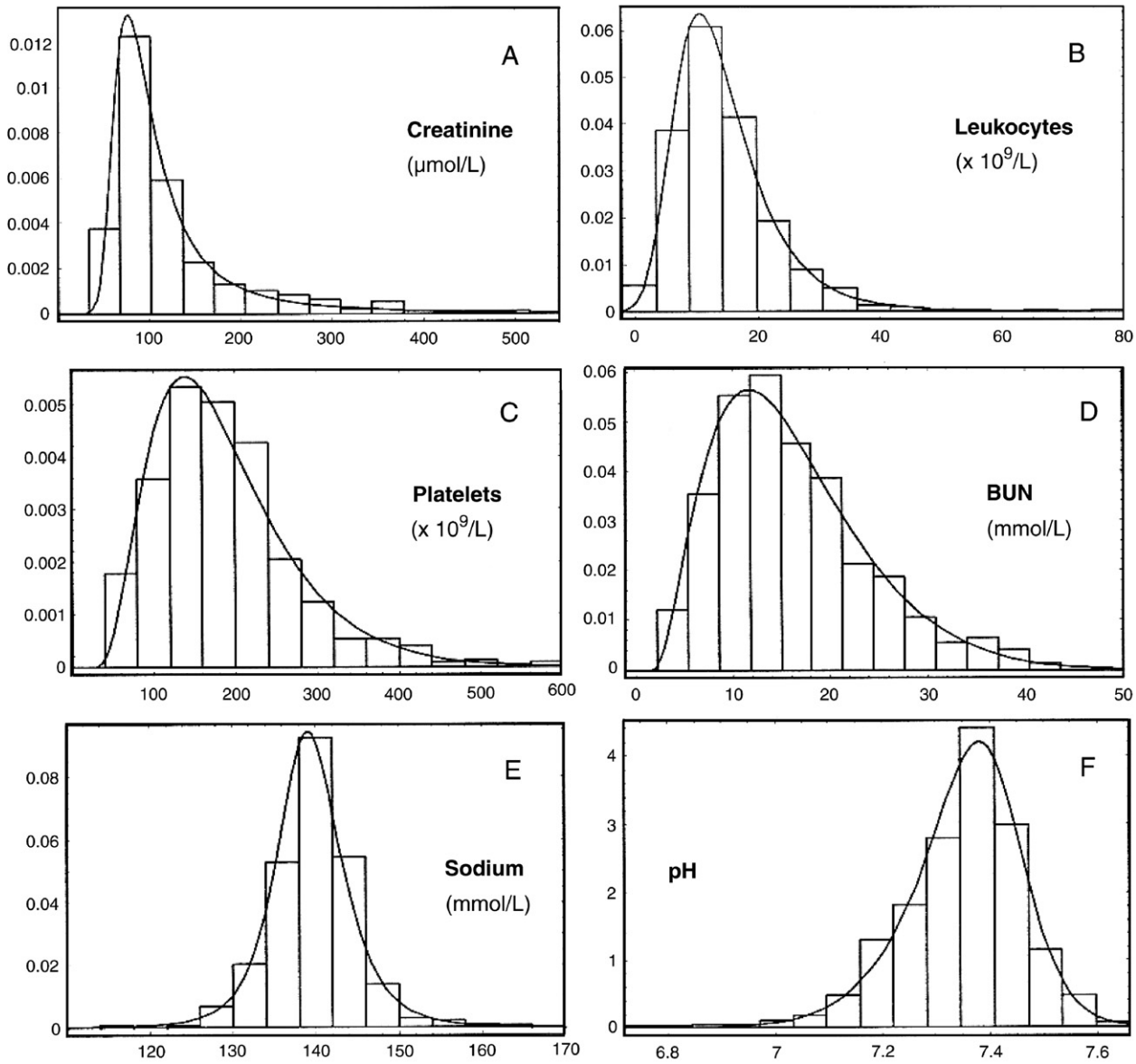

Fig. 6. GS-distributions fitted to actual data. The pdf of the fitted distribution is shown by a continuous line. (A) Creatinine (µmol/L). Fitted GSD[97.78, 26.41, 1.16, 0.015, 1.55]. (B) Leukocytes ($\times 10^9$/L). Fitted GSD[13.04, 0.99, 0.94, 0.20, 1.08]. (C) Platelets count ($\times 10^9$/L). Fitted GSD[169.79, 0.44, 0.84, 0.02, 0.94]. (D) Urea Nitrogen (BUN) (mmol/L). Fitted GSD[14.72, 0.71, 0.73, 0.13, 0.87]. (E) Serum Sodium (mmol/L). Fitted GSD[139.28, 13.40, 1.52, 0.04, 1.13]. (F) pH. Fitted GSD [7.36, 9.77, 0.88, 1.66, 0.80].

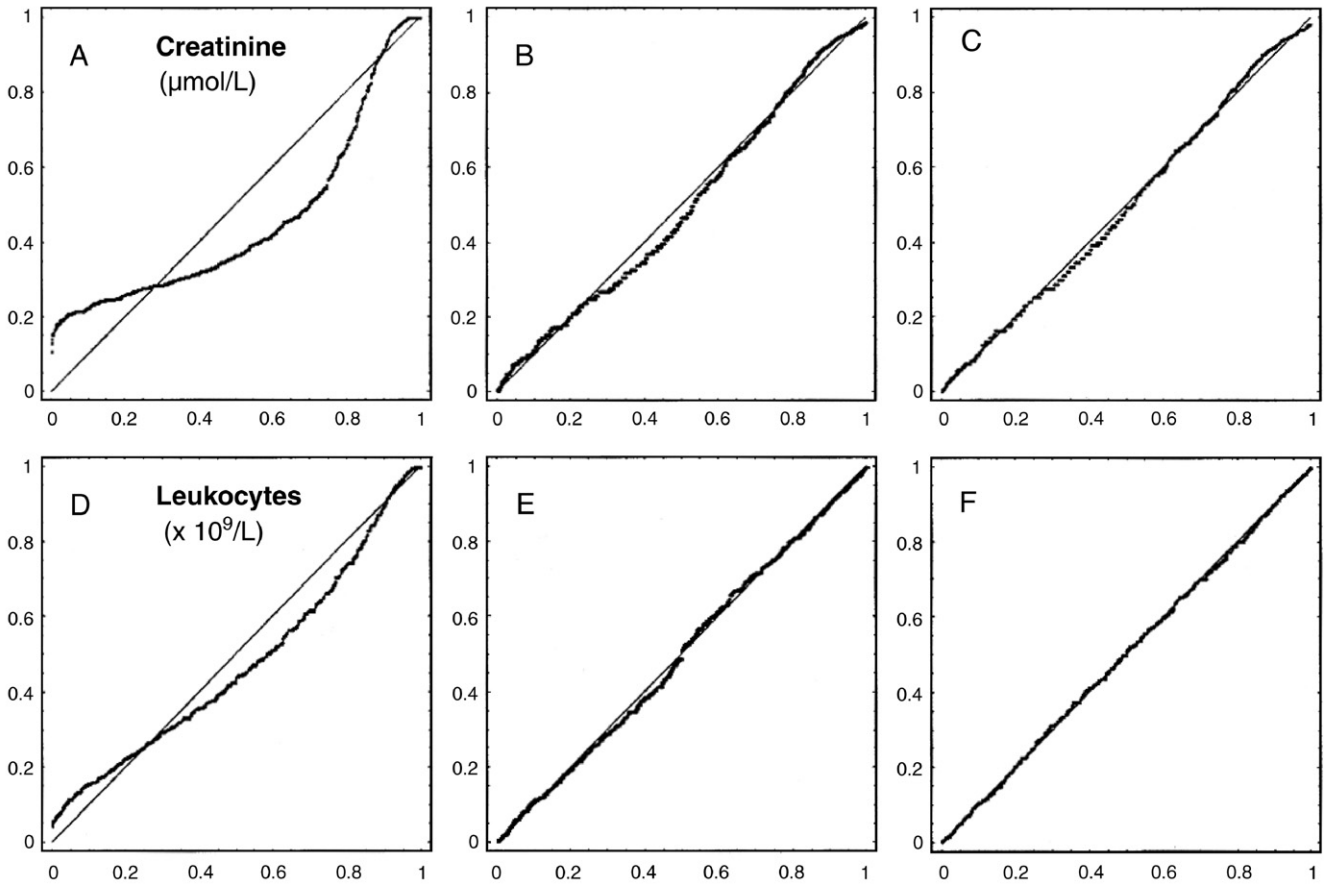

Fig. 7. *P–P* plots of data sets of Fig. 5. (A, B, C) Creatinine; (D, E, F) Leukocytes. In each case, the *P–P* plots represent: (A) and (D) Raw data compared to normal distribution. (B) and (E) Transformed data compared to normal distribution. (C) Raw data compared with the fitted GSD[97.78, 26.41, 1.16, 0.015, 1.55]. (F) Raw data compared with the fitted GSD[13.04, 0.99, 0.94, 0.20, 1.08].

etc., provide a valuable decision-making tool for clinicians and are fundamental in clinical laboratories [28–30]. The various strategies that can be used for estimating RI provides appropriate results, although do not yield an estimation of the distribution [13,31,32]. The GSD method presented in this paper introduces a new perspective. By using a general family of distributions we are able of providing a suitable estimation of the RI and a distribution model for the corresponding population. This may be an advantage when it comes to define RI in terms of a covariate [19,20]. Furthermore, once a GSD is fitted to a given data set, this can be used for complementary computations [16]. For instance, we could compute Receiver Operating Characteristic (ROC) curves by fitting a GSD to a data set of healthy subjects and to a data set of pathological subjects. Then, following the methodology already presented elsewhere [18], we can easily obtain a ROC curve.

The GSD method can provide a valuable alternative to other existing methods for RI estimation. Our results show that, at the worst, it is almost equivalent to the TST method, with the added value of providing an estimation of the underlying distribution. The GSD can be easily extended to deal with cases in which a mixture of different populations is present (work in preparation). Furthermore, GSD could be used to define hypothesis tests that are an alternative to non-parametric methods. We are investigating these possibilities to define a general framework for data modeling based on GSD.

Acknowledgements

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Appendix A. Parameter estimation in GSD

When a GSD is used as a parametric model for data one may be concern by issues regarding overparametrization. In the GSD family the number of parameters is the minimum that guarantees to include a number of distributions as particular cases in a simple function of $F(x)$. In some cases, the number of parameters can be reduced. For instance, symmetric distributions requires only three parameters, as for symmetric distributions $g=\gamma$ and $k=1$, i.e. GSD(x_0,α,g,1,g). If one knows that the underlying distribution is normal, then two parameters should suffice. In that case, the normal distribution is approximated by a GSD[x_0,α,0.8379,1,0.8379]. Thus we need to estimate only two parameters.

Table 3
95% Reference intervals (with 90% confidence intervals) for diverse variables of the data base (861 intensive care patients)

		2.5th percentile	97.5th percentile
Creatinine	NP	55.2 (51.8–57.1)	359.7 (340.1–373.4)
(μmol/L)	TST	51.2 (48.3–56.1)	370.6 (333.9–407.5)
	HDM	55.4 (52.0–56.7)	359.4 (335.9–378.8)
	GSD	53.7 (51.3–56.2)	377.3 (360.3–439.4)
Leukocytes	NP	2.80 (2.40–3.57)	35.7 (32.0–38.3)
($\times 10^9$/L)	TST	3.36 (2.97–3.76)	34.1 (32.0–36.2)
	HDM	2.96 (2.47–3.55)	35.1 (32.8–37.4)
	GSD	3.33 (2.96–3.73)	35.1 (32.7–37.4)
Platelets	NP	63.2 (61.1–66.8)	407.7 (364.6–401.3)
($\times 10^9$/L)	TST	65.8 (62.0–68.6)	383.4 (374.7–426.2)
	HDM	64.3 (61.3–67.0)	405.4 (385.1–424.8)
	GSD	66.7 (63.8–69.6)	393.3 (377.6–409.9)
Urea Nitrogen	NP	5.02 (4.64–5.42)	36.1 (34.6–37.1)
(mmol/L)	TST	5.02 (4.64–5.41)	34.3 (33.2–35.3)
	HDM	5.02 (4.65–5.42)	35.7 (34.6–36.8)
	GSD	5.01 (4.64–5.04)	34.6 (33.6–35.3)
Serum Sodium	NP	128.5 (127.5–129.6)	150.6 (148.0–152.9)
(mmol/L)	TST	128.1 (127.0–129.1)	151.6 (150.4–152.9)
	HDM	128.6 (126.7–129.4)	150.9 (148.7–153.2)
	GSD	128.2 (127.1–129.2)	150.8 (149.7–152.0)
pH	NP	7.11 (7.08–7.14)	7.55 (7.54–7.56)
	TST	7.11 (7.09–7.13)	7.54 (7.53–7.55)
	HDM	7.11 (7.08–7.13)	7.55 (7.54–7.56)
	GSD	7.11 (7.09–7.13)	7.54 (7.53–7.55)

(NP) Nonparametric approach. (TST) Two-stages transformation method. (HDM) Method of Harrel and Davis. (GSD) Method of GS-Distribution. Results are expressed as the mean and the 5th and 95th percentiles of 200 bootstrap samples in each case.

A maximum-likelihood estimator for the GSD parameters can be obtained according to the following procedure. In the case of a GSD, the likelihood can be written as:

$$L = \prod_{i=1}^{n} f(x_i) = \prod_{i=1}^{n} \alpha F(x_i)^g (1-F(x_i)^k)^\gamma \quad (6)$$

Taking logarithms, we obtain the log likelihood as:

$$\mathrm{Log}(L) = n\alpha + \sum_{i=1}^{n} g\mathrm{Log}(F(x_i)) + \sum_{i=1}^{n} \gamma \mathrm{Log}(1-F(x_i)^k) \quad (7)$$

Unfortunately, maximum-likelihood estimators cannot be obtained analytically from this expression. In that case, a numerical procedure is needed. Computation of the log likelihood requires obtaining the value of $F(x_i)$ for each sample value. This must be numerically computed by integrating Eq. (1) from x_0 to x_i. Once these values obtained, substitution into Eq. (7) produces the corresponding log likelihood for a set of parameters. Using this strategy, the parameter estimation proceeds in an iterative manner until Log(L) is maximized for a set of parameters. As an example, Figs. 2 and 6 provide some examples of fitting GSD to simulated and actual data. In all cases, the fitted distribution provides a proper representation for the data.

Confidence intervals for the GSD parameters and quantiles must be obtained by bootstrapping. For a sample data of size n boostrap samples are obtained by selecting n data points from the original sample with replacement. This is repeated a number of times to obtain a set of bootstrap samples. Then, a GSD is fitted to each of those samples. Using the different GSD so obtained, we form lists of the different parameter values and compute the desired quantiles for each distribution. The confidence intervals of each parameter and quantile are obtained by computing the 2.5 and 97.5 quantile values of each list.

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