Nonparametric Bootstrap Method for Estimating the Upper Confidence Limits of Exposure Point Concentrations of Formaldehyde in Fur Products – A Case Study of Tongxiang City, China

by

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Abstract

The distribution characteristics of the formaldehyde (FA) content of fur products produced in Tongxiang City over three consecutive years (2014-2016) were analyzed by the Lilliefors test, quarterback robust statistical method, and kernel density estimation. The bootstrap simulation sampling method was used to calculate the upper confidence limits (UCL) of the exposure point concentration (EPC) of FA. The robustness and rationality of the UCL estimation were verified by the distribution of the mean value of each bootstrap sample dataset. The results showed that their distributions were close to normal distribution but were highly skewed. The one-sided 95% UCL on the mean were 95.45 mg/kg, 93.02 mg/kg and 86.64 mg/kg, which decreased year by year. The Wilcoxon rank sum test indicated that the population distribution of the FA content in 2016 showed a structural improvement over those of the previous two years.

Introduction

The fur industry, like the leather industry, an age-old enterprise, provides a broad range of consumer goods, such as shoes, garments, bags, and so on, by turning the food industry's waste products into a desirable, useful and sustainable range of end products.[1] With the discovery of the means of removing hair, fur dressing and leather tanning took separate but parallel paths.[2] Fur is any animal skin or part thereof, with the fur fiber, hair, or fleece attached thereto either in its raw or processed state.[3] The fur processor deals with these different types of inseparable fibers, and his task is to maintain or enhance the properties of the fur fiber (keratin) first the skin fiber (collagen) second but equally.[4]

Formaldehyde (FA: CAS 50-00-0), a reactive chemical first synthesized in 1855,[5] did not find an application in fur processing until the 1920s, and its affinity for both collagen and keratin makes it an extremely useful material in tanning, pre-treatment before bleaching, dyeing, prevention of shedding of hair and so on.[6] Though FA was not directly used, most of the syntans and some chemical auxiliaries available today in the market are products made from a condensation polymerization reaction using FA as the condensing agent.[7,8] As a result, FA may be found in fur products as a result of inadvertent migration and evaporation from products during the process of tanning, finishing and using.[9,10] FA has a penetrating odor, it irritates the eyes and mucous membranes, and it has been found to have an allergenic potential.[11,12] It is classified as a probable human carcinogen by the International Agency for Research on Cancer.[13] Therefore, FA safety issues have already seriously impacted international trade and the manufacture of fur and leather products. Several stringent international regulations on the use and presence of FA in fur products have been established. In Europe, leather products need to meet the requirements that limit the content of FA (≤150 mg/kg) according to the criteria of the European Eco-label for footwear set forth in the Commission Decision 2002/231/EC.[14] In China, the National Compulsory Standard GB20400 Leather and Fur-Limit of harmful matter has a strict limit for FA in fur products. In general, FA is restricted depending on the final application and end consumer of the product. The typically acceptable limits in fur are 20 mg/kg for infant products (36 months or under) and 75 mg/kg for adult products with direct contact to skin and approximately 300 mg/kg if there is no direct contact with skin.[15]

FA contamination in fur and leather products has attracted worldwide attention in recent years.[16] Therefore, carrying out scientific and efficient risk analyses is the key to the current

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work. Obviously, estimation of the concentration of a chemical in the environment was a key element of the risk assessment process. This concentration, commonly termed the exposure point concentration (EPC), is a conservative estimate of the average chemical concentration in an environmental medium.[17] Because of the uncertainty associated with estimating the true average concentration, the upper confidence limit (UCL) of the arithmetic mean should be used for this variable.[18] The most commonly used limit is the 95% UCL, which corresponds to the 95% coverage probability of the arithmetic mean of the sample data.[19]

For the UCL calculation, the parameters method is only applicable to common data distributions, such as normal and lognormal distributions. Research indicates that a deviation occurs when the data exhibit a high variance or moderate to high skewness.[20] In this case, the nonparametric method based on computer-aided simulation techniques can effectively solve the problem. At present, these methods have played an increasingly important role in environmental exposure assessments of hazardous chemicals. Nonparametric methods deal with the existing data by use of computer-aided simulation sampling technology without consideration of the distribution of the experimental data, and it can directly obtain the desired value. It does not cause significant errors due to inappropriate assumptions of population distributions and often has a good robustness. At present, the mainstream simulation sampling methods are Bootstrap, Jackknife, and Permutation, and among these methods, Bootstrap is the most commonly used, which is recommended by international authorities.[21] Bootstrap procedures are robust nonparametric statistical methods and can be used to construct approximate confidence limits for the population mean. In these procedures, repeated samples of size *n* are drawn with replacement from a given set of observations. The process is repeated a large number of times (e.g., thousands), and each time, an estimate of the desired unknown parameter (e.g., the sample mean) is computed.[22]

In this paper, a detection data set of FA contents in fur products for three consecutive years (2014-2016) was collected and summarized. Based on replacement from a given set of observations, the bootstrap method was used to resample and the distribution of the mean value of each bootstrap sample dataset was established. A calculation and comparison of the 95% UCL for each year were performed. The Wilcoxon rank sum test was used to check the position parameters of the three-year sample data. The trend of change of FA contents in fur products over the past three years was confirmed.

Sample Sources and Methods

Case Study Area: Tongxiang City

Tongxiang City is located on the southeast shore of Taihu Lake in the northern part of Zhejiang Province and the northern region of Hang-Jia-Hu Plain.[23] It is at the center of a triangle formed by Shanghai, Hangzhou, and Suzhou (shown in Figure 1). It has three streets, nine towns, two new districts, and a population of 820,000 under its jurisdiction. Chongfu Town, one of nine towns of Tongxiang City, was awarded the "Chinese fur famous town" honorary title by the China Light Industry Federation and the China Leather Association in September 2005. The Chongfu fur industry is one of the key characteristics of the block economy in Tongxiang City, with a total of 1552 fur enterprises, more than 20000 employees and a gross value of fur industrial output of 11.48 billion yuan in 2011.[24] Therefore, it is representative and necessary to carry out the research on the estimation of the EPC of FA in fur products in the above area.

Sample Sources

The data of the FA content in fur products produced in Tongxiang City (involving hundreds of fur product manufacturers) was collected randomly by the unit to which the first author belongs: the third-party inspection-Test Center of Jiaxing Fur and Footwear Research Institute, and the monitoring continued for three years (2014-2016).

Method for Determination of FA

The FA content in fur products was measured according to the standard 'ISO 17226: Leather-Chemical determination of

Figure 1. Schematic configuration of the case study area: Tongxiang City.

formaldehyde content-Part 2: Method using colorimetric analysis' after aqueous micellar extraction of the shredded fur sample. The limit of detection (LOD) was 20 mg/kg.

Data Processing Method for Samples below the LOD

Processing methods for samples below the LOD directly affects the EPC estimation during the risk assessment of the products. There are currently three types of treatment methods for such data, including substitution, parametric methods, and nonparametric methods. The substitution method is widely used because of its simplicity and ease of understanding. According to the ratio of samples below the LOD, it was replaced by 0, 1/2LOD or LOD. GEMS/food-euro noted that if less than 60% of samples are below the LOD, it is reasonable to assign all ND samples to 1/2LOD; if more than 60% of samples are below the LOD, all ND samples should be replaced with the 0 and the LOD to generate the estimated values.[25]

Polymorphic Analysis of FA Content for Every Year

The statistical characteristics of the FA contents in 2014, 2015 and 2016 were described by using the quarterback robust statistical method. The kernel density estimation was used to describe the data polymorphism. The distribution characteristics were obtained.

Estimation UCL for EPC of FA for Every Year

Repeated sampling of polymorphic distribution data was conducted using the bootstrap method. By simulating the sampling of the original data, the UCL for the EPC of the FA content in fur products was obtained. The correlation algorithms for the data distribution, morphological analysis, UCL estimation, and bootstrap simulation sampling procedures were programmed in MATLAB 7.11(produced by the MathWorks, Inc., United States).

Pairwise Comparison of FA Content by Wilcoxon Rank Sum Test

The Wilcoxon rank sum test is a nonparametric approach to establishing a significant difference between two sample groups using magnitude-based ranks, which was a hypothesis test method.[26] The original hypothesis was that the position parameters of the two groups were consistent. If the hypothesis was established, the rank of the two groups should not differ very much. The correlation algorithm for the rank sum test was programmed in MATLAB 7.11.

Results and Discussion

Data Processing Method for Samples below the LOD

The ratios of samples below the LOD in 2014-2016 are listed in Table I. Because all three ratios were less than 60%, according to the GEMS/food-euro suggestion, the processing method was selected to assign all ND samples to 1/2LOD, and the processed data were used as the original data for the following data analysis.

Descriptive Statistical Analysis of the FA Content in Fur Products

For the normality test of the 2014-2016 data, the Lilliefors normal test was used because the number of samples was greater than 50. Under a certain confidence level (1-a=0.95), the inspection value was compared with the Lilliefors critical value to confirm the data distribution hypothesis. The Lilliefors statistics of the FA content in 2014-2016 were 0.2511, 0.2523 and 0.2766, but the critical values that fit the normal distribution were 0.0283, 0.0320 and 0.0309. It is obvious that the FA contents in 2014-2016

Table I
Processing method for samples below LOD.

Year	Sample Size	Number of below LOD	Ratio (%)	Processing method
2014	1020	271	26.57	assign all ND samples to 1/2LOD
2015	794	230	28.97	
2016	852	311	36.50	

Table II
Fundamental statistical parameters of the FA content in fur products (2014-2016).

Statistic	2014	2015	2016
Minimum (mg/kg)	10.00	10.00	10.00
Maximum (mg/kg)	843.60	921.50	1004.80
Mean (mg/kg)	87.58	84.44	78.49
First quartile (mg/kg)	10.00	10.00	10.00
Median (mg/kg)	48.90	45.25	39.70
Third quartile (mg/kg)	103.10	105.30	94.25
Variance	13365	12445	13339
Standard deviation	115.61	111.56	115.94
Coefficient of variation	1.32	1.32	1.48
Skewness	2.78	3.10	3.33

were not normally distributed. To avoid the influence of abnormal values on the level of the FA content, the statistical characteristics of FA were described using the quarterback robust statistical method. The results are shown in Table II.

Symmetry Analysis of Distribution of FA Content in Fur Products

The coefficient of skewness is a measure of the degree of symmetry in the variable distribution.[27] The skewness value can be positive, negative, or even undefined. As a general rule of thumb, if the skewness is less than -1 or greater than 1, the distribution is highly skewed. If the skewness is between -1 and -0.5 or between 0.5 and 1, the distribution is moderately skewed. If skewness is between -0.5 and 0.5, the distribution is approximately symmetric.[28] The skewness of the FA contents presented in Table III was 2.78, 3.10 and 3.33. All of them were greater than 1, so the distributions of the FA contents were highly skewed. Previous studies have shown that a deviation occurs in the UCL calculation when the data exhibit high variance and moderate to high skewness. As the skewness increases, the reliability of the calculated UCL value decreases. In such a case, nonparametric methods were recommended for the UCL computation according to EPA OSWER 9285.6-10. Kernel density estimation was used to analyze the polymorphism of the FA content in fur products. A continuous and smooth kernel density profile was constructed by processing the original discrete sample points, which was convenient to observe the distribution characteristics of the data. The kernel density distribution curves of the 2014-2016 FA contents were obtained by adjusting the default window width coefficients, as shown in Figure 2. It is observed that the distribution characteristics of the FA content were right-skewed. The right tail is longer, and the mass of the distribution is concentrated on the left of the figure.

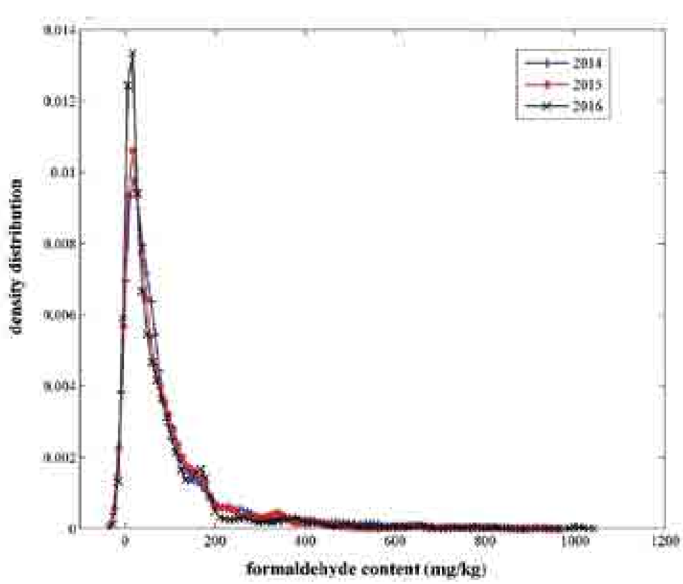

Figure 2. Probability distribution of the FA content by kernel density estimation

Computing UCL and Programmed by MATLAB

Much work has been done in recent years to develop more robust forms of bootstrapping. The bootstrap-t method is a nonparametric method that uses the bootstrap methodology to estimate quantiles of the pivotal quantity.[29] It is especially useful for skewed datasets.[30] An elaborated procedure is described as follows.

Let X_1, X_2, ..., X_n represent the n randomly sampled concentrations (a given set of observations).

STEP1: The system functions *mean (X)*, and *std (X)* are called to calculate the $\overline{X}$ and *s*.

STEP2: Write a customized function named *function1*, seeking to calculate different values of *t*, as follows:

function A= function1 (X, B) % B, repeated times, a very large number
for i=1:B, S=unidrnd (n,n,1); % construct a n×1 random matrix with a random number in 1-n
M=X(S); % generate a bootstrap sample data set

A(i)=(mean(M)-mean(X))×sqrt(n)/std(M); % compute the pivotal variable t end

STEP3: The system function *sort (A)* was called to sort *t* according to its numeric value from small to large. The MATLAB command is as follows:

t=sort (A);

Under a certain confidence level (1- α), k=B×α/2, the value of *t(k)* was calculated.

STEP4: Compute the one-sided (1-α) confidence limit on the mean.

$$UCL_{1-\alpha} = \overline{X} - t(k) \times \frac{s}{\sqrt{n}}$$

Note: *Because this value depends on random deviations, it can vary slightly on recalculation.*

Table III
UCL values of FA contents in fur products 2014-2016.

Year	2014	2015	2016
95% UCL (mg/kg)	95.45	93.02	86.64

UCL Values and Robustness Verification

The method stated above was applied to estimate the UCL with B=1,000 bootstrap iterations. The study shows that this bootstrap iteration for the vast majority of bootstrap applications has sufficient accuracy.[31] The one-sided 95% UCL of the mean is shown in Table III.

The effect of the bootstrap resampling simulation can be examined by the distribution of the mean value of each bootstrap sample dataset. If every resampling point was basically satisfied by the normal distribution, the UCL estimation could be considered to be robust. A frequency histogram and the kernel density were used to represent the density distribution of the mean value of each bootstrap sample dataset. The result is displayed in Figure 3. It is observed that the density distribution shows obvious characteristics of a single distribution. To further prove the validity of the robust estimation, the distribution

Table IV

Results of the Wilcoxon rank sum test of FA content for three consecutive years.

Groups	2014 and 2015	2015 and 2016	2014 and 2016
P value	0.5450	0.0074	0.00051
H value	0	1	1
Conclusion	acceptance	rejection	rejection

Note: H=0, which means the accepted hypothesis; H=1, which means the rejected hypothesis; P>0.05 represents the accepted hypothesis; P<0.05 indicates rejected hypothesis. (P value: return probability; and H value: return to the hypothesis test result).

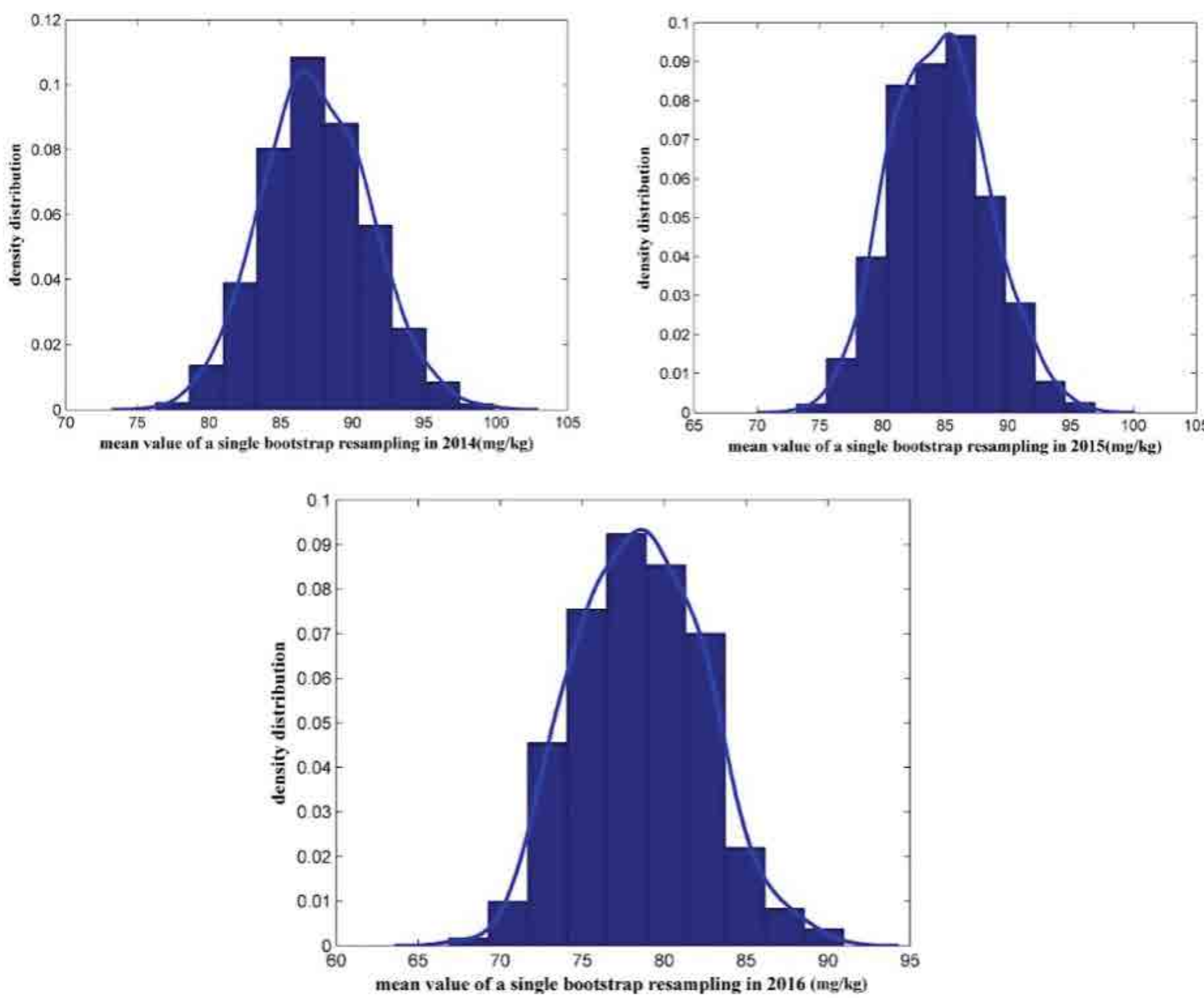

Figure 3. Density distributions of the mean value of each bootstrap sample dataset

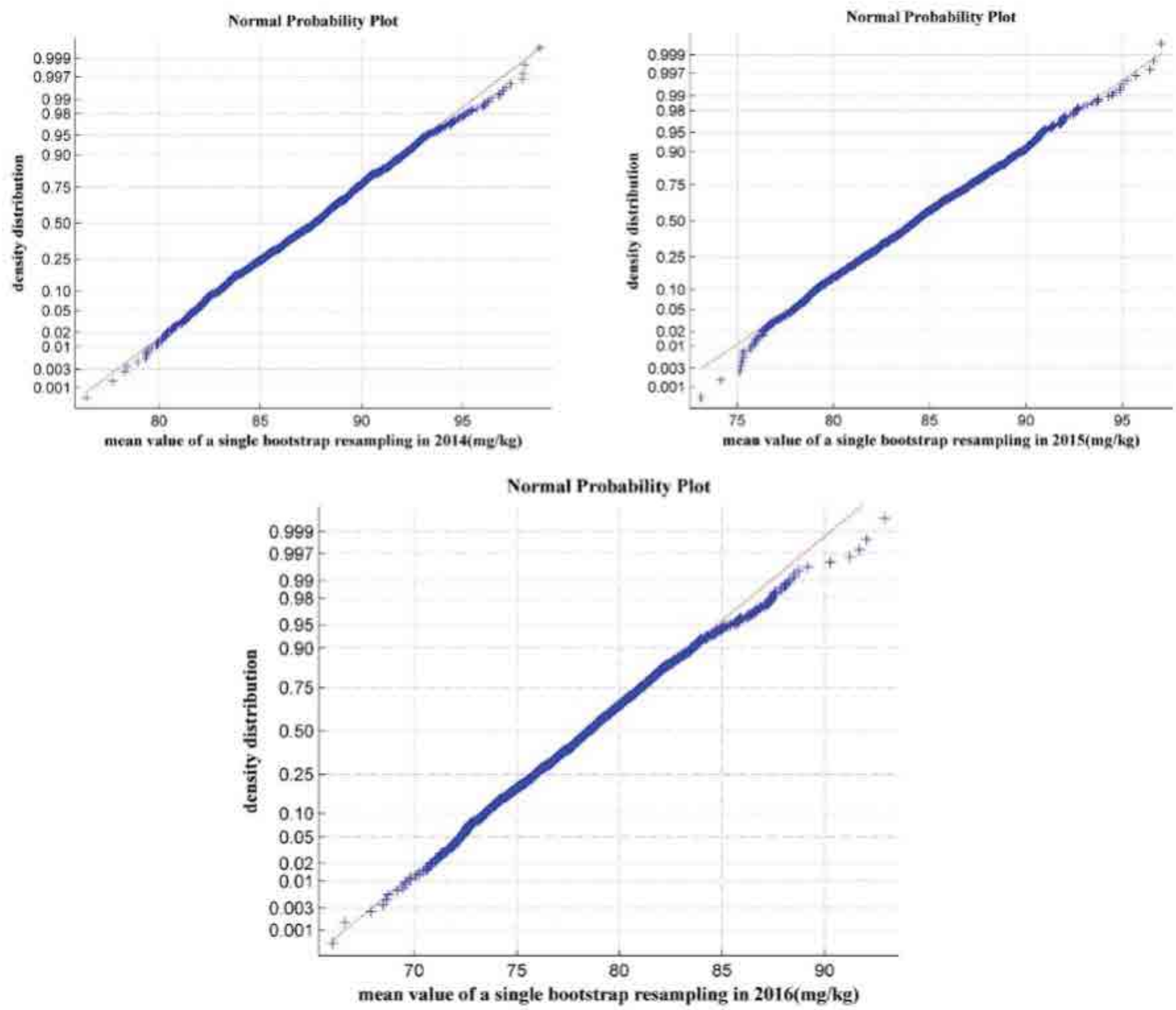

Figure 4. Normplots of the mean value of each bootstrap sample dataset

characteristics of the mean value of each bootstrap sample dataset in the normal probability distribution were investigated. The result is shown in Figure 4. It is observed that the distribution of the mean value of the bootstrap resampling was in accordance with the normal distribution, and it was proved that the UCL estimation using the bootstrap-t method was reasonable and effective.

Pairwise Comparison of FA Content by the Wilcoxon Rank Sum Test

From the results of the Lilliefors test, the data of three consecutive years does not conform to the normal distribution. Therefore, the *t*-test was not suitable, and the Wilcoxon rank sum test was adopted. The data in 2014, 2015 and 2016 were compared pairwise using MATLAB. The statistical results are shown in Table IV. The results of the comparison between the three groups showed that the population distributions of 2014 and 2015 were the same and that of 2016 was different from 2014 and 2015. At the same time, it is observed from Table II that the average and median values of the FA content in fur products decreased year by year. The results in Table IV indicated that this reduction in 2016 was a true reflection of the distribution of the sample data rather than an accidental result of random sampling. That is to say, the quality of fur products in 2016 in study area (Tongxiang City) improved structurally.

Conclusions

The distribution patterns and statistical characteristics of the FA contents in fur products for three consecutive years (2014-2016) in Tongxiang City were analyzed. The bootstrap simulation sampling method was used to calculate the UCL of the EPC of FA. The one-sided 95% UCL values of the mean were obtained, and the robustness and rationality of the UCL estimation were verified. The results showed that the UCL values from 2014-2016 decreased gradually, which showed that the risk level of EPC in fur products decreased year by year. Particularly, in 2016, the population distribution of the FA content showed a structural improvement over the previous two years. This method overcame the defects of a non-normal distribution and small sample size. It can be universally applied for unknown data distribution morphologies and provides a new method for feature analysis of detection data. It is useful for performing risk assessments of FA in fur products in follow-up studies.

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