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Proper sampling, total measurement uncertainty, variographic analysis & fit-for-purpose acceptance levels for pharmaceutical mixing monitoring

K.H. Esbensen[a,b] and R.J. Romañach[c]
[a]Geological Survey of Denmark and Greenland (GEUS). Copenhagen. Denmark. E-mail: ke@geus.dk
[b]ACABS research group, Department of Chemistry and Bioscience, University of Aalborg, campus Esbjerg (AAUE), Denmark
[c]University of Puerto Rico, Department of Chemistry, Mayagüez, PR. E-mail: rodolfoj.romanach@upr.edu

Process monitoring in technology and industry in general, in pharmaceutical batch and continuous manufacturing in particular, is *incomplete* without full understanding of all sources of variation. Pharmaceutical mixture heterogeneity interacts with the particular sampling process involved (by physical extraction or by Process Analytical Technology (PAT) signal acquisition) potentially creating four Incorrect Sampling Errors (ISE), two Correct Sampling Errors (CSE) in addition to the Total Analytical Error (TAE). In the highly regulated pharmaceutical production context it is essential to eliminate, or reduce maximally, all unnecessary contributions to the Total Sampling Error (TSE) to the Measurement Uncertainty (MU_{total}) in order to be able to meet stringent regulatory blend and dose uniformity requirements. Current problems mainly stem from inadequate understanding of the challenges regarding sampling of powder blends. In this endeavor the Theory of Sampling (TOS) forms the only reliable scientific framework from which to seek resolution. We here present the variographic approach with an aim to conduct TSE error variance identification and to show how to develop *fit-for-purpose* acceptance levels in critical powder blending process monitoring. The key issue regards the nugget effect, which contains all non-optimised [ISE, CSE] plus TAE contributions to MU_{total}. A large nugget effect w.r.t. the sill is a warning that the measurement system is far from fit-for-purpose, and must be improved. Regulatory guidances have hitherto called for physical sampling from within blenders, leading to significant ISE associated with the insertion of sample thieves (sampling spears). Instead of self-crippling spear sampling we here call for a paradigm shift, very much from the TOS regimen, in the form of alternative on-line variographic characterisation of 1-D blender outflow streams. Practical illustrations and case histories are described in parallel contributions to WCSB7.

Introduction

Process monitoring in technology and industry in general, in pharmaceutical batch and continuous manufacturing in particular, is incomplete without full understanding of all sources of variation. Pharmaceutical mixture heterogeneity interacts with the particular sampling process involved, either by physical extraction or by PAT signal acquisition, potentially creating four Incorrect Sampling Errors (ISE), two Correct Sampling errors (CSE), and two process sampling errors (PSE) – in addition to the analytical error (TAE). In the highly regulated pharmaceutical production context it is essential to eliminate, or reduce maximally, all unnecessary contributions to the total measurement uncertainty MU_{total} when developing scientifically justifiable monitoring procedures. For the present overview, focus is on the effectiveness of mixer blending which is the last active processing step before tableting, i.e. how can it be ascertained that a particular blend has reached a mixing level that complies with the required 'homogeneity' and uniformity limits. The specific pharmaceutical manufacturing background was introduced to the TOS community by Romañach & Esbensen.[1] TOS provides the necessary tools to separate sampling errors from process variation, critically needed for full understanding of all sources of the sum-total of process, sampling and analytical variation.

Heterogeneity – also at the endpoint of mixing

Blending of fine-grained powders may be considered at both macro and micro-mixing scales. The proportion of a single Active Pharmaceutical Ingredient (API) may, or may not, be well distributed throughout the blend. The blend also includes other components, called excipients, that are important for various reasons. The blending process seeks to break up drug aggregates present at the beginning of the process. However, there is always a possibility that some aggregates will not respond completely if they are mainly located in an "inactive" location within the blender. Sampling methods have been developed to try to target material from such "dead spots" with an aim to protect patients from a potential drug overdose. Thus, differences in drug distribution within blends have been extensively investigated in the pharmaceutical industry, using a wide variety of analytical techniques (but largely without proper understanding of the associated sampling errors effects), and all have shown a significant scale-hierarchy of blend heterogeneity, ranging from a single dose (e.g. tablet) to the entire blender volume (mg-g-kg realm).

Heterogeneity in the framework of TOS focus on the central notion that all types of materials are heterogeneous at two fundamentally different scales, which gives rise to the two essential features: Constitutional Heterogeneity (CH) (heterogeneity between the fundamental compositional units) and Distributional Heterogeneity (DH) (heterogeneity between all virtual sampling increments throughout the lot). In the pharmaceutical realm, the focus has been to achieve "homogeneity" after the blending process (e.g. an API and several excipients) is completed. The term "homogeneous" is here not used to indicate when all units making up the lot are identical (TOS' definition), but refers to a blend with an acceptable low level of drug distribution variability, i.e. a fit-for-purpose homogeneity. The acceptable threshold drug distribution variability has been a

relative standard deviation (RSD) of less than 5% in many contexts. It is worth noting that this is identical to the demand in material balance operations, but considerably lower than requirement for commercial sampling (1%).

When a sampling process interacts with a lot with a specific heterogeneity, two sampling errors arise, the Fundamental Sampling Error (FSE) and the Grouping and Segregation Error (GSE) which influences the total MU. This is of course a trivial concept in TOS, but not in pharma: it bears noting that the differentiation into CH and DH is virtually unknown here, which is one of the reasons that a fully comprehensive theory of mixing has been very long in the making (50-60 years), and first is beginning to show a final conceptualization in the two first decades after the millennium. Sampling errors have been recognized in pharmaceutical studies, although not characterized in the same way and to the same level of comprehension as within TOS. A recently withdrawn guidance on sampling of powder blends indicated: "Sampling errors may occur in some powder blends, sampling devices, and techniques that make it impractical to evaluate adequacy of mix using only the blend data. In such cases, we recommend that you use in-process unit data in conjunction with blend sample data to evaluate blend uniformity."[2] The same document also indicated that: "If blend sampling error is detected, more sophisticated, statistical analyses should be applied to assess the situation".

However, such statistical evaluations are post fact, complex and do not give indications of how to eliminate the causative problem(s). The best approach, in pharma as everywhere else in science, technology and industry, is to completely avoid unnecessary and controllable sampling errors in the monitoring of manufacturing processes in the first place as stipulated by TOS. We here outline a radical way out of the blender sampling predicament in pharma, which amounts to a paradigm shift with respect to the current traditional situation.

Theory of mixing – does it help reducing MU_{total}?

A mixing theory is all very well – but does it help in reducing the adverse effects of sampling errors, the latter a notion that has just begun to be acknowledged in the pharmaceutical realm? The history of the evolution of a theory of mixing is presented elsewhere; only a few key aspects are necessary for the present overview.

1) It has always been assumed that effective mixing will lead to a perfect *random mixture*, and most theoretical analysis has been carried out on this background. This has a serious impact on how to address real-world mixing end-products however. It turns out that this is not a realistic end-point understanding (see further below regarding residual heterogeneity).
2) A very influential misunderstanding is that there has been only very little recognition that sampling processes suffer from significant errors inflicted by the processes themselves, i.e. Incorrect Sampling Errors (ISE). The one notable exception is that of Muzzio et al.,[3] which analysed in considerable detail the effects of using thieves for sampling of pharmaceutical mixtures, and which must be credited for pointing out the highly adverse effects resulting from forcing thieves through an in-homogenous medium ('clumped', segregated, layered) as well as casting a first empirical light on differential flow characteristics for API's and excipients respectively. API's and excipients are often of significantly different crystal/particle size and forms which can lead to markedly different flowability with resulting different mixability consequences, significantly hindering terminal mixing efforts.

How to sample from *within* a container – that is the question!

Pharmaceutical companies are extracting powder mixtures directly from blenders to check blend uniformity, and this is almost universally carried out using sampling thieves (sampling spears).

Figure 1 shows the recommended approach for what is currently considered to be adequate sampling from a V-blender. Note that

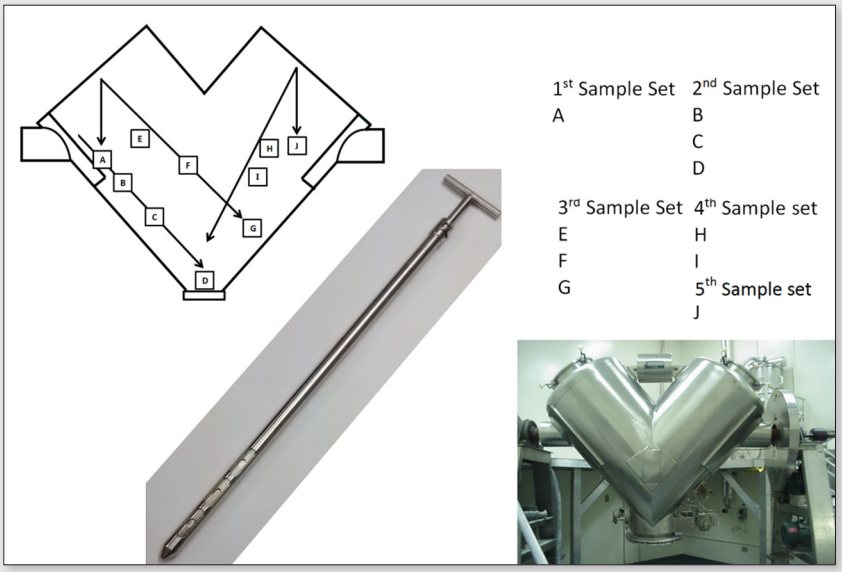

Figure 1. Traditional thief sampling (spear sampling) from *within* pharmaceutical mixing blenders (here a tumbling V-blender) recommends using 10 fixed locations organised in a certain order intended to minimize 'drag down' of powder from higher locations.[4] The fundamental assumption is that these locations (including replication at a few locations) represent the "most in-homogenous" parts of *any* blend, for *all* types of mixtures, in *all* types of blenders. Alas this assumption is untenable in the industrial practice.

each sample obtained from this geometrical scheme is analysed individually, there is specifically no requirement to aggregate these 10 singular samples into composite samples, because the objective is to estimate the residual heterogeneity present *after* mixing. This scheme is therefore forcing what is fundamentally a grab sampling approach, which has resulted in numerous difficulties w.r.t. the accuracy and precision of the desired quality check of the final blended mixtures. The sample thieves employ small, pre-set cavities to assure that the samples extracted has approximately the mass of a single dose unit, which from a 'consumer' point of view is a reasonable demand and a cogent solution: the analytical result must pertain to the dose unit the patient receives. The operation of pharma sampling thieves is otherwise standard: the cavities are closed when the metal rod is inserted into the blender and first opened for powder to flow into the cavity when reaching the appropriate location in the blender, and then closed again to remove the extracted material from the specific location targeted. However such a small sample size unavoidably forces the attending FSE to be at a maximum.

It is on this basis that a recommended geometrical set of fixed locations is assumed to be able to render a reliable quantification of the residual heterogeneity in the entire blender volume. From a TOS perspective, this is clearly an unsustainable assumption however. Sample thieves are unable to furnish representative samples under almost all circumstances – except regarding exceedingly uniform mixtures, which is of only little help when trying to monitor an ongoing mixing process, or trying to verify whether a mixing endpoint satisfies a regulation threshold, i.e. most of the times this sampling approach is used, the mixture will not be at its lowest heterogeneity near 'uniformity'. The fixed geometrical scheme sets the order in which the mixture is to be sampled with an aim to minimize the effect of disturbance of the powder bed (N.B. not to eliminate, but only to minimize this disturbance). Thief sampling is not an easy task in practice since blenders are quite large and accessibility is often restricted in the industrial practice. Thus, typically only 6 to 10 grab samples are removed from blenders following only minor variations of the master plan as illustrated, Figure 1.[4] Also, recent publications indicate that regulatory agencies want to understand the local sample-sample variation at specific locations, e.g. Reference 5. Multiple insertions of the sample thief at a specific, or a few pre-selected location(s) will only complicate the evaluation of mixing – this is just more disturbance of the final product caused by biased sampling unit operations.

Any set of fixed locations will not be able to target the worst "hidden zones" that is supposed to be associated with maximum variations in drug concentration in a comparable manner - for *all* types of compositionally different mixtures, for the *range* of different dimensions in current industrial and experimental blenders (very serious scale-up issues abound). TOS' Fundamental Sampling Principle (FSP) is systematically broken in all fixed location sampling plans, e.g. six fixed locations,[6] or 10 fixed locations in the conventional V-blender geometry,[4,7] resulting in a virtual certainty for non-representative sampling, DS 3077.[8] Thief sampling is very nearly always unable to deliver "correct sampling" in practice, which forces one to accept a sampling bias, as has been demonstrated in many practical studies in the TOS realm and also within pharma.[3] But no sampling bias can ever be estimated, nor can it ever be corrected for - with any means. In other words, the current paradigm in pharma is structurally and fatally flawed.

This state of affairs is critically serious but may not necessarily be unavoidable – TOS to the fore.

The starting task is therefore to discontinue efforts to demonstrate the adverse effects from biased sampling processes; the objective is directly the opposite: to embark on a program with an aim to eliminate all bias-prone sampling procedures, equipment and programs within pharma, i.e. to eliminate all that has to do with ISE. TOS offers a suite of practical solutions on how to eliminate or reduce the effects from the full complement of sampling errors [ISE, CSE] not in need to be iterated in detail here, suffice to point to References 9–12 and further references herein.

An iconoclastic solution – Do not sample from *within* a container!

We here propose a radical way out of the current situation in pharma - do *not* sample from within blenders!

All mixing products (with or without sampling-for-quality control) will eventually be discharged from the mixing container and transported to the tableting/encapsulating equipment immediately upon termination of the mixing stage. This process will unavoidably add to the material heterogeneity due to an assured impact of flow-segregation (pouring segregation); it is only a matter of how much additional flow/pouring segregation is heaped upon the carefully mixed product. This added heterogeneity will not be observed, or accounted for, until quality control of the final product units (tablets, capsules), i.e. any such heterogeneity is left unobserved. If the final product variability is found to be exceeding the pertinent regulatory threshold the whole batch will have to be discarded. It would have been far better if this case had been established *before* tableting and packaging, i.e. *en route* to the tableting unit, preferentially just before this last unit operation commences.

Romañach & Esbensen indicate an alternative, indeed optimal quality inspection location is on the blender output stream (obviously after the added outflow segregation impact).[1] For the sake of argument, picture the flow en route to the tableting unit as a mini conveyor belt, or similar.[13–16] The argument is that the length extension of this flow is a *linear mapping of the entire container volume* now allowing complete insight into the residual material heterogeneity after termination of mixing (plus whatever level of added flow-age segregation variability) – in stark contrast to today's situation characterized by the impossibility of adequate sampling from within the blender.

This proposal is a simple rectification that eliminates all errors associated with sampling thieves while acknowledging that the mixture is always also impacted by some level of segregation upon leaving the blender vessel. Blender sampling is to be discontinued and replaced by on-line process sampling of the output stream at a suitable location. With TOS competence, it is an easy matter to establish an effective, un-biased sampling and/or PAT signal acquisition situation on a flowing stream of matter with a small cross-sectional dimension and thus reap the full benefits of process sampling.[9–11,17,18]

Variographic characterisation of mixing processes – a new twist

Perhaps the most important issue in current pharmaceutical blending is: How to be able to recognize, identify and estimate the magnitude of the sum-total of sampling + analytical error effects influencing the total Measurement Uncertainty (MU_{total}) in current system

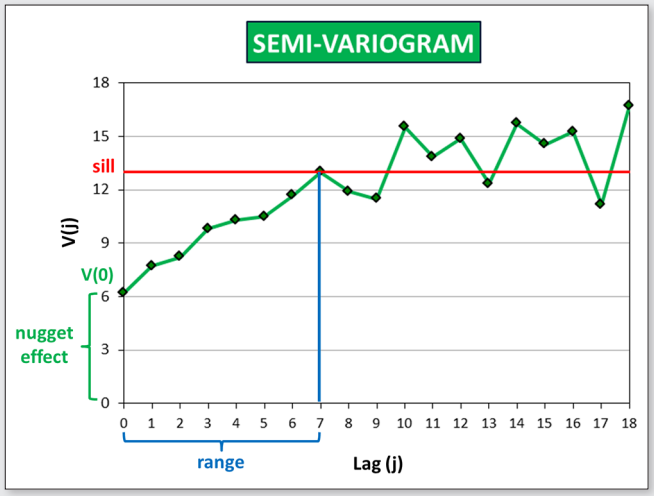

Figure 2. Generic variogram characterized by its three principal parameters: range, sill and nugget effect

implementations? TOS shows that there are many opportunities for process monitoring through the use of variographic analysis a.o. providing estimates of the nugget effect (n.e.) and the sill (MU_{total}).[8–12,17] The only necessary-and-sufficient condition is to be able to set up a TOS-correct variographic experiment, a task that will be easy to perform in the well regulated manufacturing and processing environments in pharma. N.B. All variograhic characterisation must be based on unbiased sampling processes and data (see further below).

All variograms are characterised by three principal parameters: the range, the sill and the nugget effect.

A powerful TOS insight concerns the variogram nugget effect as the magnitude made up of the sum-total of all sampling and analysis error effects contributing to the MU_{total}, i.e. [TAE, CSE, ISE]. Thus the degree to which efforts have not been fully successful in either eliminating the incorrect sampling errors, or reduce them optimally [leaving only CSE], will unavoidably show up as factors increasing the magnitude of the nugget effect.

TOS outlines that the nugget effect variance can also be viewed as the Minimum Possible Error (MPE), and how it is always possible, in principle as well as in practice, to reduce MPE either by sampling at an increased rate and/or by compositing more increments. If/when MPE is found to be "high", this is a sign that the current measurement system is marred by unacceptably high error contributions and that something must be done about it.

While these facts regarding the variogram are well-known in the TOS realm, they are virtually unknown in pharma! There is here a very fertile opportunity to introduce variographic analysis.

The variogram monitors the mixing process and at the same time characterizes the measurement system. Regarding the latter objective, it is only necessary to relate the nugget effect to the sill both as estimated by the experimental variogram. The sampling standard, DS 3077 (2013) a.o. established a generic measurement system quality index, termed $RSV_{1\text{-}dim}$, defined as the n.e./sill (expressed as a %-age). The smaller the $RSV_{1\text{-}dim}$ index, the better the measurement system will allow insight into the true process variation, as unencumbered by MU_{total} as possible.

Figure 3 shows the principal difference between an acceptable measurement system $RSV_{1\text{-}dim}$ ~30% (while appearing high this measurement system will still be able to "see" all pertinent process/product variations) and its unacceptable counterpart ($RSV_{1\text{-}dim}$ > 85%) as revealed by these simple variogram characteristics.[8]

For a perfectly mixed material, the variogram must appear flat. Any vestiges of imperfect mixing will be revealed by the form and level of the sill of the output variogram. Any significantly irregular sill 'morphology' will signify less than perfect mixing. The more a variogram represents the final state of a well-mixed blend, the smaller the overall sill.

It is never an issue to ascertain significant deviation from a flat variogram; neither is locating the lowest sill level, as shown in Figure 4 where, as an example, four alternative mixing processes variants are compared. Note that even for the lowest of the four variograms

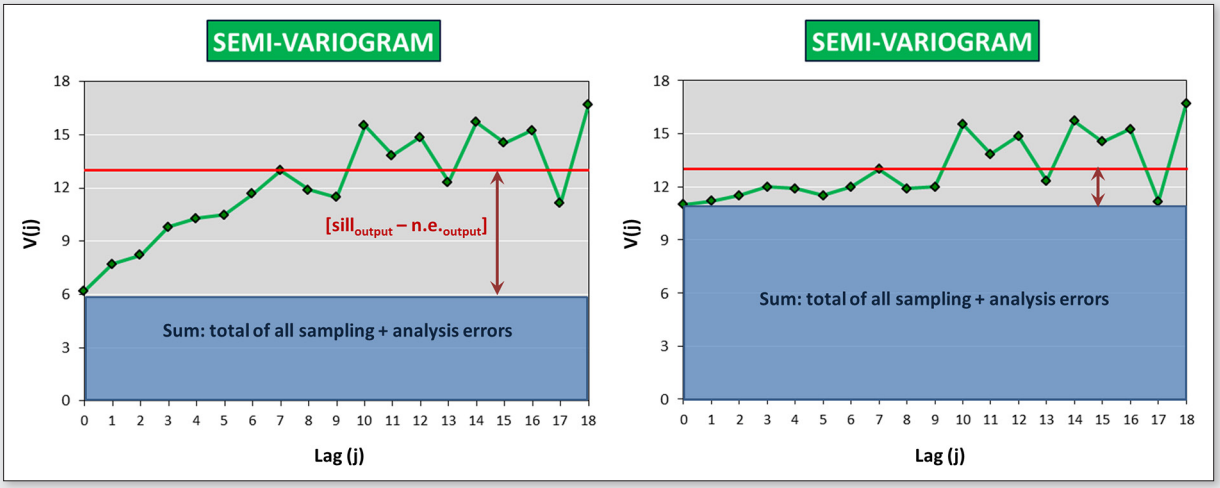

Figure 3. Principal difference between an acceptable measurement system (left) and its unacceptable counterpart (right); $RSV_{1\text{-}D}$ is ~30% (left), but >85% (right). The situation illustrated represents variographic analysis of a pharmaceutical blender output streams with identical sill levels for comparison, i.e. with similar total process variability. Resolving adverse sampling issues (reducing the nugget effect) may result in a significantly lower overall sill as well, see Figure 4.

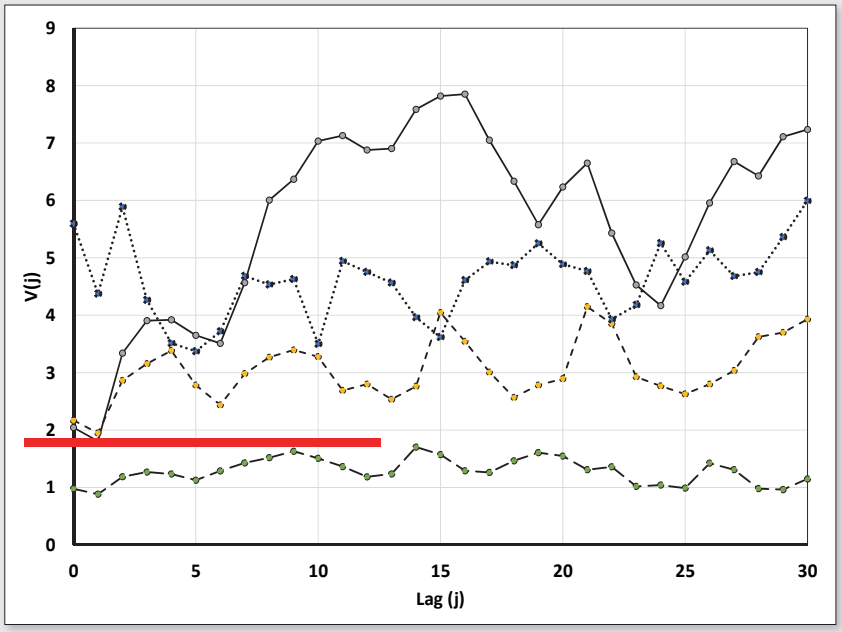

Figure 4. Schematic illustration of variograms of four alternative mixing process variants in pharmaceutical formulation development. The process represented by the bottom variogram is optimal because of its lowest sill level and least deviations from a flat variogram. All variograms reveal one form or other of feeder periodicity inheritance, only sufficiently dampened in the bottom one. Note regulator threshold criterion (horizontal line). Even though the optimal variogram is not flat the fact that it falls exclusively below the regulator threshold allows the blending process to be declared *fit-for-purpose*.

there is a minor, residual deviation for intermediate lags. Figure 4 also shows how the variograms relate to a regulator threshold translated into a variance level. As soon as when the sill is below the threshold, the mixing/blending process can be declared "fit-for-purpose".

For a blender output variogram (indeed for all process variograms) the 'true' process variation, i.e. the effective material residual variability after termination of mixing, is not the sill itself but the *corrected* sill: $sill_{output} - n.e._{output}$, arrived at by subtracting the effective MU_{total}. Thus a flat variogram does not necessarily signify a perfect, ideal mixture. Non-zero corrected sill levels: $sill_{output} - n.e._{output}$ represents *residual mixture heterogeneity* which never vanishes completely for all naturally occurring or technological mixtures, e.g. Reference 19. Thus it is the flat, low-level variogram with a non-zero corrected sill: $sill_{output} - n.e._{output}$ that represents the realistic, real-world end-point of all mixing processes.

Once embarking on a process using variographic characterization, the road is open, also for pharma, for progressing rapidly to be able to make use of the more advanced facilities, e.g. complete identification, decomposition and estimation of all process variance contributions, V(0), V(1), V(cyclic),V(trend), e.g. Pitard.[10]

Discussion

It would appear that current Federal Drug Administration (USA) demands, which has led to extensive thief sampling, to a large degree is in contradiction to its own objectives. The bias incurred by thief sampling will always cover up a non-trivial fraction (perhaps a significant, or a fatally large) fraction of the product heterogeneity manifestation (or process heterogeneity), thus effectively disallowing it to be validly observed and interpreted.

In the case of the critical pharmaceutical blending process this is an unacceptable situation. What is needed is guaranteed full observability giving optimal possibility for critical compliance testing.

Esbensen & Romañach are currently developing the variogram approach for pharmaceutical mixing quality control directed at the blender output stream in full detail.[16,20] Focus is both on the overall sill level as well as on the corrected sill: silloutput – n.e.output. This opens up for addressing regulator threshold compliance based on a dynamic, self-correcting measurement system. When a blender output variogram lies below this threshold, e.g. Figure 4, the blending product can be declared fit-for-purpose, which is all that is needed in the given regulation context. It is then not necessary to carry on with further mixing – the product is verified ready for tableting.

In the situation where it has been demonstrated that no further heterogeneity is added during tableting, variographic characterization of the blender output stream may in fact be all that is needed in order to prove to the regulator's satisfaction that also the dual final product inspection is in fact already tested and found acceptable.[21]

The proposed variographic outflow approach provides a clear alternative to current and other proposed methods that involve sampling from within the blender.[22]

The authors are in the process of outlining the present new concept in an official whitepaper format.

For the record: all valid variographic analysis must be carried out on unbiased process data. TOS is replete with warnings, elucidations and solutions regarding this stipulation.[8–12,17–19]

Conclusion—a call for a paradigm shift

There are many opportunities for TOS to be involved in significant TSE improvements in pharma, notably w.r.t. eliminating sampling bias in the primary blender sampling stage. It is here proposed to introduce a systematic variographic approach on blender outflow streams for determining the characteristics of both the product and the monitoring system itself, whether based on physical sampling

or on on-line PAT analysers. All that is needed is the availability of relevant blender output data. Variography is a highly favourable alternative to today's practice because of its self-checking MU_{total} features, i.e. the $RSV_{1\text{-}dim}$ [%] quality index, and because it can be based on routine monitoring outflow data which can be obtained as part of the on-line manufacturing process monitoring anyway.

For measurement systems in which a successful effort has been made to eliminate ISE, i.e. unbiased systems, the nugget effect (MPE) is a reliable estimate of the remaining MU_{total} precision. In the situation where the bias issue has not been fully resolved, an increased nugget effect compared to the sill is a critical and reliable warning of an inferior or a degraded measurement system. Even in this case the corrected sill: $sill_{output}$ – $n.e._{output}$ may still be able to characterize the mixing end-result although with decreased fidelity as this difference shrinks (for worse and worse total measurement systems).

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References

1. R. J. Romañach and K. H. Esbensen, "Sampling in pharmaceutical manufacturing - Many opportunities to improve today's practice through the Theory of Sampling (TOS).", *TOS Forum* **4,** 4-5 (2015). doi: 10.1255/tosf.37
2. Guidance for Industry Powder Blends and Finished Dosage Units-Stratified In-Process Dosage Unit Sampling and Assessment, (2003)
3. F. J. Muzzio, P. Robinson, C. Wightman and D. Brone, "Sampling practices in powder blending", *International Journal of Pharmaceutics*. **155,** 153-178 (1997). 10.1016/s0378-5173(97)04865-5
4. S. Bozzone, "Solid Oral Dosage Forms Powder Blending ", *IKEV Meeting Presentation*. (2001).
5. J. S. Bergum, J. K. Prescott, R. W. Tekwani, T. P. Garcia, J. Clark and W. Brown, "Current Events in Blend and Content Uniformity", *Pharmaceutical Engineering*. **34,** 28-39 (2014).
6. J. Berman and J. A. Planchard, "Blend Uniformity and Unit Dose Sampling", *Drug Development and Industrial Pharmacy*. **21,** 1257-1283 (1995). doi: 10.3109/03639049509063017
7. R. C. Hwang, M. K. Gemoules and D. K. Ramlose, "A Systematic Approach for Optimizing the Blending Process of a Direct-Compression Tablet Formulation", *Pharmaceutical Technology*. **22,** 158-170 (1998).
8. DS 3077, "Representative sampling - Horizontal Standard". Danish Standards Foundation, (2013)
9. P. Gy, *Sampling for Analytical Purposes*, 1st. Ed. Wiley, New York (1998) ISBN 0-471-97956-2
10. F. F. Pitard, *Pierre Gy's Sampling Theory and Sampling Practice. Heterogeneity, Sampling Correctness, and Statistical Process Control*, CRC Press, (1993) ISBN 0-8493-8917-8
11. K. H. Esbensen and L. P. Julius, "Representative Sampling, Data Quality, Validation - A Necessary Trinity in Chemometrics", *Comprehensive Chemometrics: Chemical and Biochemical Data Analysis, Vols 1-4*. C1-C20 (2009).
12. K. H. Esbensen & P.Minkkinen (Eds), "Special issue: 50 years of Pierre Gy's theory of Sampling - Proceedings: First World Conference on Sampling and Blending (WCSB1) - Tutorials on Sampling: Theory and Practice", *Chemometrics and Intelligent Laboratory Systems*. **74,** 1-1 (2004). 10.1016/j.chemolab.2004.07.001
13. A. U. Vanarase, M. Alcalà, J. I. Jerez Rozo, F. J. Muzzio and R. J. Romañach, "Real-time monitoring of drug concentration in a continuous powder mixing process using NIR spectroscopy", *Chemical Engineering Science*. **65,** 5728-5733 (2010). http://dx.doi.org/10.1016/j.ces.2010.01.036
14. M. Popo, S. Romero-Torres, C. Conde and R. J. Romanach, "Blend uniformity analysis using stream sampling and near infrared spectroscopy", *AAPS PharmSciTech*. **3,** E24 (2002). 10.1208/pt030324
15. Y. Colón, M. Florian, D. Acevedo, R. Méndez and R. Romañach, "Near Infrared Method Development for a Continuous Manufacturing Blending Process", *Journal of Pharmaceutical Innovation*. **9,** 291-301 (2014). 10.1007/s12247-014-9194-1
16. R. J. Romañach and K. H. Esbensen, "Estimating total sampling error for near infrared spectroscopic analysis of pharmaceutical blends—theory of sampling to the rescue", in Esbensen, K.H. & Wagner, C. (Eds), *Proceedings: 7th World Conference on Sampling and Blending (WCSB7)* pp 71–75 (2015). *TOS forum Special Issue* IM Publications (2015). doi: 10.1255/tosf.66
17. K. H. Esbensen and P. Paasch-Mortensen, "Theory of Sampling - The missing link in PAT" in Bakeev, K. (Ed.) *Process Analytical Technology,* chap. 3. (John Wiley & Sons, Ltd, 2010) 37-80 ISBN 978-0-470-72207-7
18. F. F. Pitard, *Pierre Gy's Theory of Sampling and C.O. Ingarell's Poission process approach, pathways to representative sampling and appropriate industrial standards* (Dr. Techn. thesis), Aalborg University,Campus Esbjerg, Denmark, 2009, ISBN 978-87-7606-032-9
19. F. F. Pitard and D. Francois-Bongarcon, "Demystifying the Fundamental Sampling Error and the Grouping and Segregation Error for Practitioners", in Alfaro, M, Magri, E, Pitard, F (Eds) *Proceedings: 5.th World Conference on Sampling and Blending*, pp 39-56 GECAMIN Publ. (2011) ISBN 978-956-8504-59-5
20. A. Sánchez Paternina, A. Roman Ospino, B. Alvarado, K. H. Esbensen and R. J. Romanach, "When "homogeneity" is expected – Theory of Sampling in pharmaceutical manufacturing", in Esbensen, K.H. & Wagner, C. (Eds) *Proceedings: 7th World Conference on Sampling and Blending (WCSB7)* pp 67–70 (2015). *TOS forum Special Issue* IM Publications. doi: 10.1255/tosf.61
21. J. Bergum, T. Parks, J. Prescott, R. Tejwani, J. Clark, W. Brown, F. Muzzio, S. Patel and C. Hoiberg, "Assessment of Blend and Content Uniformity. Technical Discussion of Sampling Plans and Application of ASTM E2709/E2810", *Journal of Pharmaceutical Innovation*. **10,** 84-97 (2015). 10.1007/s12247-014-9208-z
22. T. Garcia, J. Bergum, J. Prescott, R. Tejwani, T. Parks, J. Clark, W. Brown, F. Muzzio, S. Patel and C. Hoiberg, "Recommendations for the Assessment of Blend and Content Uniformity: Modifications to Withdrawn FDA Draft Stratified Sampling Guidance", *Journal of Pharmaceutical Innovation*. **10,** 76-83 (2015). 10.1007/s12247-014-9207-0