Abstract One of the indispensable pieces of information a compounder needs with every pharmaceutical ingredient chemical is the Certificate of Analysis. In fact, it is needed with every different lot of the material, since variations from lot to lot in chemicals can be significant. This article briefly explains what a Certificate of Analysis contains, how to use it in compounding, and provides several examples to illustrate areas where errors in compounding could occur if it were not used.

William J. Zolner, PhD

In testing compounds at Eagle Analytical, we find that one of the most significant reasons for out of specification (OOS) preparations is because pharmacists do not consult the Certificate of Analysis (CoA) when compounding. While we most frequently see this in sterile preparations, it can also contribute to the OOS of nonsterile preparations. Therefore, the first recommendation I always make when speaking to a new compounding class is to insist on a CoA for each and every chemical that they receive into their pharmacy. Some compounding software actually allows the user to load information from the CoA into the software inventory database; the correcting calculations explained below can be made automatically in a pharmacy’s compounding worksheet.

What is a Certificate of Analysis?

In its simplest form, a CoA, which is a written agreement between the seller of a product and the purchaser, lists and confirms the content of selected ingredients in a product. Consumers deal with a CoA every time they purchase a can of soup or a loaf of bread at the supermarket. The next time you purchase something at the supermarket, look at the label and you will see in clear print a list of the ingredients of the item you just purchased. In this case, the government has decided which ingredients are important to list and confirm to the purchaser information such as: calories per serving, fat content, sugars, and carbohydrates. You, as a purchaser, can now use the CoA to decide between two alternate products using this information (e.g., choosing the product with the lowest fat content).

The Certificate of Analysis in the Pharmaceutical Industry

Similarly, the CoA for an active pharmaceutical ingredient (API) is an agreement between the seller of the chemical and the purchaser, warranting the content of the chemical in certain key areas. While the CoA received for an API chemical is often considerably more detailed and specific than that of a common grocery item, its function is basically the same, to let the purchaser know exactly what they have purchased. So if the CoA is an agreement between the purchaser and seller, who sets the specifications that will be met by the chemical? If you, as the purchaser, were sourcing a very
large quantity of the chemical, you could negotiate with the manufacturer to get the exact specification you want. The specification could be tailored exactly for your application and would only be limited by the physical and basic chemical constraints of the API, and, of course, the price you would be willing to pay. However, a pharmacist, even a large volume compounding pharmacist, usually does not use enough of any chemical to be able to independently set the specifications. This is where the United States Pharmacopeia (USP) comes into the picture. Working with the pharmaceutical industry, the U.S. Food and Drug Administration (FDA), and the manufacturer of a drug using the chemical, the USP proposes a set of specifications for the chemical to be used in the pharmaceutical industry. After an extensive review consultation with all parties, these specifications, along with the methods that will be used to measure or test for these attributes, are published in a USP monograph. The monograph then provides a list of specifications that a manufacturer can use as the basis for the CoA. If, along with other requirements, the chemical meets the specifications set forth in the monograph, it can be labeled with the USP designation.

As an aside, understanding how the specifications for a particular active ingredient is determined, subsequently published, and used as the basis for a CoA, can be very important to a compounding pharmacist. If the active is to be used in an unforeseen manner, as in a new dosage form, the specifications that would be critical for that use may not be covered in a standard USP derived CoA. In this case, it may be important for the purchaser to request additional tests from their supplier, over and above the standard tests, to assure that the active is applicable for their specific use.

So if the chemical that I purchase is a United States Pharmacopeia (USP) chemical, with specification attributes previously determined, why do I even need to be concerned with having the CoA? Two basic reasons:

1. While the specifications are stated, the acceptable limits can often be quite broad, especially with the water content of the chemical (as covered below). The chemical may meet the CoA requirements, but still result in an OOS final drug preparation if the actual specifications are not considered.
2. The CoA for each chemical is lot number specific. Two different lots of the same basic chemical can sometimes have significantly different actual specifications, even though both fall within the overall USP standards.

Most suppliers of API chemicals will provide a CoA for each specific lot of supplied chemical. A good standard operating procedure is to request that the CoA be provided with each shipment. At the same time, it is a good idea to also request a material safety data sheet (MSDS) for each chemical that you have in your pharmacy. I recommend that you have a hard copy (or maybe on a tablet computer) of the MSDS in your pharmacy for easy access in case you need it during an emergency situation when your computer may be inoperable.

Four Key Specifications in the Certificate of Analysis

In most CoA documents, the data is presented in a three-column format. The first listing is the specification name, the second the limits that the chemical should fall within for that specification, and the third the results of the tests on the specific lot of chemical relative to this CoA. While the CoA contains a host of information and specifications, there are four specific areas that should always be consulted.

1. The NAME of the chemical and its Chemical Abstracts Service (CAS) number: Every chemical is given a unique (CAS) number that specifically identifies its chemical formula with the exception of polymers. Sometimes people get sloppy with names, abbreviations, base, or salt
forms or just get the name wrong, which is why the CAS Number is so valuable. If you know the CAS number, you know the chemical. The CAS number and the name of the chemical have a one-to-one correspondence. If in doubt about the name of the chemical, you can look up the CAS number in several reference texts or on the Web to make certain that the CofA is naming your chemical correctly. This is particularly important in those instances where a salt form of the chemical may be confused with the base or for chemicals with water of hydration in the formula. For example, fentanyl (437-38-7) is considerably different from fentanyl citrate (990-73-8); and magnesium sulfate (7487-88-9), magnesium sulfate trihydrate (15320-30-6), and magnesium sulfate heptahydrate (10034-99-8) are all different.

2. The DESCRIPTION of the chemical: The description of a chemical is an explanation of what the chemical looks like, if it has a characteristic odor, and if it exhibits any identifying unique observable characteristics. Your human senses of sight, smell, and observation are surprisingly keen in determining variations and abnormalities in how things should look and smell. If you open a vial of chemical and it is a fine white powder with black specs, but the CofA just says it should be a fine white powder, you know immediately that this should be concern for questioning the chemical. In a similar manner, a foul smell for something that should have a faint citrus odor is an instant tip off that the chemical may need to be set aside and not used.

3. The ASSAY of the chemical: The assay of the chemical is the amount of the pure API in the material that you have, or in some cases, as with antibiotics, the biological activity per weight of the material. This may also be expressed in activity units, as with hyaluronidase (723 IU/mg) where the chemical has been compared to an industry standard and assigned an International Unit designated assay or strength. One thing to understand is that in most of the chemicals the assay is expressed on a dry or anhydrous basis (that is after the water has been removed from the material). So an API with an assay of 98.0% and a water content (see below) of 7.0% really only contains 910 mg/g of the pure API. The other area where the assay can be confusing is in those cases (infrequent) where the assay refers to another form of the named API. As an example, for the API amikacin sulfate, the assay refers to the amount of amikacin base in the amikacin sulfate. Unfortunately, this subtle distinction is often left off the description of the assay in many CofAs, so the only way one would know is to refer back to the API monograph in the USP.

4. The fourth area for attention is the designation specifying the amount of WATER in the chemical: This is in most cases referred to as water, where the Water is water of hydration (chemically bound to the molecule) or as Loss on Drying (LOD) where the water is absorbed onto the material. For example, a specification for Water for or an LOD of 6.3% means that when you weigh out 100 mg of the material (6.3 mg is really water). Also, keep in mind that a high LOD means that the chemical has a propensity to absorb water, and the LOD percentage is the water that was present at the time of analysis, usually not too long after manufacture. Chemicals that are improperly stored, or in geographic locations that may have high humidity conditions, may continue to absorb water if not carefully handled, resulting in significantly under-potent APIs.

There are other items on the CofA that may also be important from an informational point of view, such as manufacture date, expiry date, and molecular weight of the chemical. There may also be specifications that are important to your particular use of the chemical or API that are not covered by the CofA. I am reminded of a situation several years ago where patients undergoing chelation therapy with EDTA disodium were excreting large amounts of aluminum. This was traced to the EDTA disodium chemical. It was a USP chemical, but while there is a specification for heavy metals in the monograph, there is no corresponding specification for aluminum. However, once this was known, suppliers began testing the EDTA disodium for aluminum to make sure it was suitable for this application. So, while the CofA is important, it is still necessary to critically look at your specific application and evaluate the CofA on those specifications that are critical for your unique use.

How Do I Correct For Base/Salt, Assay, and Water in My Compound?

Now that a pharmacist knows the exact API they have, its assay, and the amount of water (as Water or LOD), let’s discuss how this information can be used when compounding a preparation.

1. Base/Salt Corrections

The first thing when considering Base/Salt corrections is to determine what the prescription is requesting. Unfortunately, many prescribers do not fully appreciate the subtle and sometimes not so subtle differences between the exact name of an API and the casual name. A good example is lidocaine hydrochloride (HCl), which many times is used and referred to as lidocaine. There is a 23% lidocaine concentration spread between two preparations made with the same amount of these APIs. Other APIs may have a more significant outcome, such as fentanyl and fentanyl citrate, which have a 57% difference in concentration for the same amount of material. Even more confusing is when the name of the preparation does not correspond to the salt form of the API. For example, leucovorin calcium injection calls for a potency based on just the leucovorin, whereas the API assay is based on leucovorin calcium. I could continue here with examples, but that would only confuse the situation. However, faced with this, what is a compounder supposed to do? First, consult your prescriber if in doubt as to what they want. It may also be a good idea to first check with the USP to see if a monograph is available and have this information ready when you consult the prescriber. If there still is doubt, by all means label the preparation correctly. For example, do not call a preparation lidocaine 1% if you made it with 1% of the hydrochloride salt; call it lidocaine HCl 1%.

Now that the pharmacist knows how the preparation is to be prepared (base or salt), how does he correct for the proper weight of the chemical he has? For this, the pharmacist will need to obtain the molecular weights of the base and salt form of the...
chemical. The ratio of these two molecular weights will provide the correction factor to apply.

Example
The ratio of the molecular weights (MW) of fentanyl (MW 336.47) and fentanyl citrate (MW 528.59) indicates that it takes 1.5710 mg of fentanyl citrate to yield 1.000 mg of fentanyl. Therefore, if the prescription calls for 50 mcg/mL of fentanyl, 78.55 mcg/mL of fentanyl citrate needs to be made. In a similar manner, if 1% lidocaine (MW 234.34) is needed, 1.23% lidocaine HCl (MW 288.81) is needed.

2. Assay Correction
The correction for the assay content of the API can be calculated if the assay is given as a percentage, for example 98.3%. This means that the API has only 0.983 g of API per g of chemical. Many antibiotic CoA documents give this number directly, for example 983 mg/g (0.983 g/g). Dividing the weight required in the preparation by this ratio will provide the amount of the chemical to weigh out for an accurate preparation. If the assay is in units, you may need to calculate directly the weight of the chemical necessary, as with hyaluronidase. For example, if I am compounding 100 mL of 150 U/mL, and I have an assay of 725 U/mg, I will need 20.69 mg of the chemical.

3. Water Correction
Water (or LOD) is usually expressed as a percentage content on the CoA, such as 9.6% water. This means that proportionally every 100 mg of the chemical actually has 9.6 mg of water. Another way to look at this is that the amount of API in the sample is actually [(100-Water%)/100] g/g chemical. Therefore, in a similar manner to the assay, the weight of pure API that needed should be divided by this ratio to determine the actual weight of the chemical needed for this preparation.

Just to further confuse matters, there are preparations that do not require a correction for the salt form or water content of the chemical. The most notable being morphine sulfate injection, which is made with morphine sulfate pentahydrate, and the potency of the injection is based on morphine sulfate pentahydrate. Again, when in doubt consult the USP for an applicable monograph.

Putting It All Together
Since all of these corrections will most likely need to be done for the same preparation, and since multiplication is cumulative, these corrections can be applied in any order or in one step.

Example
How much chemical would be needed to make 100 mL of a 10-mg/mL leucovorin calcium injection with an API of leucovorin calcium where the CoA lists the assay as 96.5% and the water content as 12.2%?
From the monograph for leucovorin calcium injection, potency is determined by just the base, leucovorin, so a base/salt correction will need to be made. The MW of leucovorin calcium is 511.50 and the MW of leucovorin is 471.42, so 1.0850 mg of chemical will be needed for every mg of leucovorin.

The assay of the API, measured on a dry basis, is 96.5% or 0.965 g/g of chemical. The LOD is listed as 12.2%, which means that there are only 0.878 g of API/g of chemical.

Needed: 100 mL × 10 mg/mL = 1000 mg of leucovorin
= 1000 mg leucovorin × salt/base correction/assay correction/water correction
= 1000 mg × (1/0.965) × (1/0.878) = 1280 mg of chemical

Therefore, 1280 mg of chemical is needed to make the preparation.

Conclusion
As shown from the above discussion, the use of the CoA of an API chemical in compounding can be very important. The difficulty is that there are many exceptions to the rules, so giving a simple explanation and developing an encompassing procedure for their use is a daunting undertaking. Knowing that this can be confusing, what can a compounder do to minimize the risk of making an OOS preparation? Several suggestions are as follows:

• A pharmacist should make sure she knows what the prescriber wants in the preparation. If the prescriber specifies a concentration relating to the base chemical and a salt is being used, the pharmacist will need to account for the salt content in the preparation.
• Always consult the CoA for the preparation’s API. Since the error band for potency is usually +/- 10% in compounded preparations, if the assay is close to 100% and water content is small, it may be acceptable to ignore the corrections.
• Use a trusted source for preparation formulas. If you cannot locate a reputable source for a formula, consult a colleague who you know may be compounding the preparation and ask them to offer assistance.
• It is amazing how willing respected compounding pharmacists are to share their information.
• Obtain a copy of the USP. I know it is expensive, but it has a wealth of information and answers many questions that daily arise in the pharmacy.
• Test your preparations with a third-party quality-control testing laboratory to make sure that you are compounding a “quality” preparation.

Address correspondence to William J. Zolner, PhD, President, Eagle Analytical Services, 9881 South Wilcrest Drive, Houston, TX 77099. E-mail: bzolner@eagleanalytical.com