



# BEYOND PESTICIDES

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**Docket ID # AMS-NOP-20-0089**

## **Re. CS: Kasugamycin Petition**

These comments to the National Organic Standards Board (NOSB) on its Spring 2021 agenda are submitted on behalf of Beyond Pesticides. Founded in 1981 as a national, grassroots, membership organization that represents community-based organizations and a range of people seeking to bridge the interests of consumers, farmers and farmworkers, Beyond Pesticides advances improved protections from pesticides and alternative pest management strategies that reduce or eliminate a reliance on pesticides. Our membership and network span the 50 states and the world.

Kasugamycin is an antibiotic, and as such, the petition should be denied. Now that we have learned what a pandemic looks and feels like, with the astounding levels of infection, hospitalization, and death from COVID-19, we must take serious steps to prevent another pandemic on the horizon—this one tied to bacterial resistance to antibiotics. An important article in *The Lancet*<sup>1</sup> points to a “looming potential pandemic” resulting from a “rise in multidrug-resistant bacterial infections that are undetected, underdiagnosed, and increasingly untreatable, [which] threatens the health of people in the USA and globally.” When streptomycin and tetracycline were presented for their final votes by the Crops Subcommittee, the committee was unanimous that the antibiotics needed to go—the question was how fast. How fast could growers get over their dependence on these antibiotics that pose threats to human health and the environment and are unpopular with organic consumers? Your predecessors on the NOSB struggled long and hard to erase the stigma of antibiotic use in organic fruit production—something that was left over from the transition of so many

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<sup>1</sup> Strathdee, S.A., Davies, S.C. and Marcelin, J.R., 2020. Confronting antimicrobial resistance beyond the COVID-19 pandemic and the 2020 US election. *The Lancet*, 396(10257), pp.1050-1053.  
<https://www.thelancet.com/action/showPdf?pii=S0140-6736%2820%2932063-8>.

chemical-intensive fruit orchards after the Alar “scare.”<sup>2</sup> Do we now want to step on that treadmill again? The reasons for rejecting the kasugamycin petition are the same as the reasons for eliminating streptomycin and tetracycline, and many will be repeated below. NOSB members who are not convinced by this reasoning should read the views presented in the dockets and transcripts of NOSB meetings from 2006 through 2014 concerning those other antibiotics. The CS proposal document on tetracycline in 2013 contains a history—mostly of efforts to remove the antibiotic.

### **Kasugamycin poses a toxicological threat.**

A Japanese risk assessment found that major adverse effects of kasugamycin are “decreased body weight gain, ulcer and others in the rectum and anus, effects on the lingua (disappearance of the papillary epithelial cells in dogs), brown pigmentation in proximal tubular epithelial cells and others in the kidney, and tubular atrophy and others in the testes. In a two-generation reproductive toxicity study in rats, the incidence of testicular abnormalities such as tubular atrophy increased in F1 parental animals, and decrease in conception rate and others were observed.”<sup>3</sup>

Kasugamycin is systemic and translocates through the plant.<sup>4</sup> While the Technical Review (TR) discusses residues on the fruit and calculates residues at harvest based on an exponential decay rate of the chemical when exposed to sun and air, it does not address residues in plant tissues, including fruit. The parent compound (kasugamycin per se) was the major identified component in all samples from all harvest intervals.<sup>5</sup>

The TR states, “Accidental poisonings—assumedly via ingestion—have apparently occurred because PubChem lists emergency detox procedures. Poisonings cause respiratory distress and pulmonary edema. Seizures may also occur, which are treated with diazepam. Hypovolemia is treated with Ringers solution. If kasugamycin solutions make contact with the eyes, they are to be treated with saline.”<sup>6</sup>

### **Our lack of knowledge requires a precautionary approach.**

The complete mode of action of a pesticide is not always known when it is registered—especially non-target effects. Take the example of the fungicide fludioxinil, a phenylpyrrole fungicide, which was developed to treat seeds during storage, but which has come to be used commonly on grains, vegetables, fruits, and ornamental plants during cultivation, and to treat

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<sup>2</sup> Shistar, T, 2013. A is for Apples, Alar, and Antibiotics. *Pesticides and You*.

<https://www.beyondpesticides.org/assets/media/documents/infoservices/pesticidesandyou/documents/AisforApplesCited.pdf>.

<sup>3</sup> Food Safety Commission of Japan, 2014. Risk Assessment Report: Kasugamycin. English excerpt:

[https://www.fsc.go.jp/english/evaluationreports/pesticide/kasugamycin\\_fs246.pdf](https://www.fsc.go.jp/english/evaluationreports/pesticide/kasugamycin_fs246.pdf).

<sup>4</sup> Advertisement for Kasu-B at <https://www.dhanuka.com/details/kasu-b>.

<sup>5</sup> USEPA, Health Effects Division; Human Health Risk Assessment for Proposed Food Uses of the Fungicide Kasugamycin on Importing Fruiting Vegetables (Group 8) (August 2005). Available from, as of November 9, 2006: <http://www.regulations.gov/fdmpublic/component/main>, from PubChem.

<sup>6</sup> Kasugamycin TR, lines 723-726.

produce after it has been harvest.<sup>7</sup> In its coverage of a study published in March 2019, the American Association for the Advancement of Science publication, EurekAlert,<sup>8</sup> reported that, “The ability of [the fungicide] fludioxonil to act on a sugar-metabolizing enzyme common to all cells, and to produce the damaging compound methylglyoxal, may mean that the pesticide has more potential to harm non-fungal cells than previously thought. Although fludioxonil has been deemed safe for use, the authors suggest that the effects of this widely used pesticide has upon animals be re-examined.”

Although fludioxonil is effective in killing fungi, the mode, or mechanism, of action for this pesticide was previously not well understood. In a previous investigation,<sup>9</sup> Drs. Brandhorst and Klein pointed to the uncertainty about how fludioxonil actually causes fungal cell death, asserting that this uncertainty merits a reevaluation by the U.S. Environmental Protection Agency (EPA) of its potential impacts on human health, noting reports of the fungicide’s ability to disrupt liver, endocrine, and neurological systems. Prior to this current study, it was believed that fludioxonil targets hybrid histidine kinase (HHK), a protein in fungal cells. Regarding the mechanism of action, Syngenta theorized that fludioxonil binds to HHK, activating a biochemical process that causes fungal cells to kill themselves. In 2016, Dr. Klein’s lab team found that, although fludioxonil needs HHK in order to kill fungi, the pesticide and protein do not directly interact.

The scientists in Dr. Klein’s lab turned to the hypothesis that oxidative stress—a common effect of pesticides on their targets— an imbalance between cells that are oxygen-producing free radicals and antioxidant defenses in the body—might be the linchpin. The team found that when they exposed fungi to various kinds of oxidative stress, cells remained healthy, but that fludioxonil inhibits an enzyme related to cellular sugar metabolism, causing (via a spike in methylglyoxal release) activation of the deadly HHK cascade.<sup>10</sup>

Dr. Brandhorst notes, “The take home lesson is that fludioxonil is multifactorial. It is not compromising cells by one solitary mechanism. It has potential to damage cells in a variety of ways.” He references a 2007 investigation that demonstrated that, in fungi, disruption of glutathione homeostasis (which manages oxidative stress) synergistically enhances the toxicity of fludioxonil, suggesting that an oxidative stress response pathway may overshadow osmoregulation functions. Fludioxonil has been shown to have DNA damaging impacts on human liver cells, and Dr. Brandhorst suspects that depletion of the intracellular antioxidant glutathione may ultimately be identified as a factor in fludioxonil-related liver damage. The

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<sup>7</sup> See a longer version of this story in: Simes, D, 2020. Highly Destructive Pesticide Effects Unregulated. *Pesticides and You*, Winter 2019-2020.

<sup>8</sup> AAAS, 2019. New mechanism of action found for agricultural pesticide fludioxonil. [https://www.eurekalert.org/pub\\_releases/2019-03/uow-nmo032119.php](https://www.eurekalert.org/pub_releases/2019-03/uow-nmo032119.php).

<sup>9</sup> Brandhorst, T.T. and Klein, B.S., 2019. Uncertainty surrounding the mechanism and safety of the post-harvest fungicide fludioxonil. *Food and Chemical Toxicology*, 123, pp.561-565.

<sup>10</sup> Brandhorst, T.T., Kean, I.R., Lawry, S.M., Wiesner, D.L. and Klein, B.S., 2019. Phenylpyrrole fungicides act on triosephosphate isomerase to induce methylglyoxal stress and alter hybrid histidine kinase activity. *Scientific reports*, 9(1), pp.1-17.

enzyme-suppressing action of fludioxonil on an enzyme common to all cells is at the heart of the alarm this research is raising, but it is not the only reason the fungicide needs to be reevaluated.

Adding to the concern about fludioxonil's mechanism of action and the implications for all organisms, including humans, is its synergistic potential. A 2012 study by French researchers found that a mixture of fludioxonil and cyprodinil, another fungicide, yields data suggesting cytotoxic (lethal to cells) and genotoxic (damaging to DNA) effects at low concentrations, with a significantly higher effect of the mixture than would be expected from an exposure response to the individual fungicides. This study by Dr. Brandhorst, et al. adds to the growing body of research on the interactive effects of pesticides on human health and the environment. Beyond Pesticides advocates for a far more precautionary approach, based on this statement on the Precautionary Principle (known as the Wingspread Statement):

When an activity raises threats of harm to human health or the environment, precautionary measures should be taken even if some cause and effect relationships are not fully established scientifically. In this context the proponent of an activity, rather than the public, should bear the burden of proof. The process of applying the precautionary principle must be open, informed and democratic and must include potentially affected parties. It must also involve an examination of the full range of alternatives, including no action.<sup>11</sup>

### **Antibiotic resistance in human pathogens is a serious public health threat, and use of antibiotics in the orchard contributes to that threat.**

The discussion of antibiotics in organic production is not new. In 2013 and 2014, the NOSB voted to eliminate the use of tetracycline and streptomycin. In the final proposal of 2014, the Crops Subcommittee acknowledged, "Both sides agree that the 'core' issue here is whether or not there is a risk of enhancing antibiotic resistance in human pathogens."

Antibiotic resistance poses a serious threat to human health. We have all had either firsthand or secondhand experience with antibiotic resistant infections —whether it is a child with ear infections that fail to respond to one antibiotic after another, a relative who died from methicillin-resistant *Staphylococcus aureus* (MRSA), someone who acquired a multiply resistant infection in the hospital, or another experience with persistent and non-responsive infections. The Infectious Disease Society of America (IDSA) estimates the annual cost of antibiotic-resistant infections to be 21 to 34 billion dollars, and states, "Just one organism, methicillin-resistant *Staphylococcus aureus* (MRSA), kills more Americans every year than emphysema, HIV/AIDS, Parkinson's disease, and homicide combined."<sup>12</sup> Thus, according to

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<sup>11</sup> Wingspread statement.1998. <https://www.sehn.org/sehn/wingspread-conference-on-the-precautionary-principle>.

<sup>12</sup> IDSA, "Facts About Antibiotic Resistance"

[http://www.idsociety.org/IDSA/Site\\_Map/Topics\\_of\\_Interest/Antimicrobial\\_Resistance/Public\\_Policy/Facts\\_about\\_Antibiotic\\_Resistance.aspx](http://www.idsociety.org/IDSA/Site_Map/Topics_of_Interest/Antimicrobial_Resistance/Public_Policy/Facts_about_Antibiotic_Resistance.aspx) accessed 2/14/2013.

IDSA, “Antimicrobial resistance is recognized as one of the greatest threats to human health worldwide.”<sup>13</sup>

### **Kasugamycin’s persistence allows time for resistance to develop.**

Kasugamycin is persistent—that is, resistant to degradation—in the soil and anaerobic aquatic and sediment conditions.<sup>14</sup> In this respect, it is similar to tetracycline<sup>15</sup> and streptomycin.<sup>16</sup> In addition, kasugamycin is systemic and translocates through the plant.<sup>17</sup> While the Technical Review (TR) discusses residues on the fruit and calculates residues at harvest based on an exponential decay rate of the chemical when exposed to sun and air, it does not address residues in plant tissues, including fruit. The parent compound (kasugamycin per se) was the major identified component in all fruit samples from all harvest intervals.<sup>18</sup> Thus, kasugamycin is present for a substantial period of time, allowing for exposure to bacteria in the orchard and consumers.

### **Kasugamycin has medical and veterinary uses.**

Kasugamycin was tested and proven to be effective against *Pseudomonas aeruginosa* urinary infections in humans.<sup>19</sup> Kasugamycin targets an enzyme important in the pathogenesis of COVID-19.<sup>20</sup> Kasugamycin can reduce the development of resistance to the anti-tuberculosis drug rifampicin and may itself prove to be an effective drug itself in fighting tuberculosis.<sup>21</sup> It has been used for animal infections.<sup>22</sup> Although it is not essential to our argument that kasugamycin itself is of medical and veterinary importance, the need for effective antibiotics is an important reason for avoiding agricultural uses that can lead to resistance in potentially valuable antibiotics.

### **Antibiotic resistance in the orchard**

In April 2013, the NOSB discussed the problem of antibiotic resistance thoroughly with respect to tetracycline and heard from numerous commenters concerning the problem of

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<sup>13</sup> IDSA, “Facts About Antibiotic Resistance.”

<sup>14</sup> Kasugamycin TR, lines 257-316.

<sup>15</sup> Tetracycline TR, 2006. Lines 261-266

<sup>16</sup> Streptomycin TR, 2011. Lines 217-225.

<sup>17</sup> Advertisement for Kasu-B at <https://www.dhanuka.com/details/kasu-b>.

<sup>18</sup> USEPA, Health Effects Division; Human Health Risk Assessment for Proposed Food Uses of the Fungicide Kasugamycin on Importing Fruiting Vegetables (Group 8) (August 2005). Available from, as of November 9, 2006: <http://www.regulations.gov/fdmspublic/component/main>, from PubChem.

<sup>19</sup> <https://pubchem.ncbi.nlm.nih.gov/compound/kasugamycin#section=Drug-and-Medication-Information>.

<sup>20</sup> Kamle, S., Ma, B., He, C.H., Akosman, B., Zhou, Y., Lee, C.M., El-Deiry, W.S., Huntington, K., Liang, O., Machan, J. and Kang, M.J., Chitinase 3-like-1 is a Therapeutic Target That Mediates the Effects of Aging in COVID-19. *bioRxiv*, pp.2021-01.

<sup>21</sup> Chaudhuri, S., Li, L., Zimmerman, M., Chen, Y., Chen, Y.X., Toosky, M.N., Gardner, M., Pan, M., Li, Y.Y., Kawaji, Q. and Zhu, J.H., 2018. Kasugamycin potentiates rifampicin and limits emergence of resistance in Mycobacterium tuberculosis by specifically decreasing mycobacterial mistranslation. *Elife*, 7, p.e36782.

<sup>22</sup> <https://pubchem.ncbi.nlm.nih.gov/compound/kasugamycin#section=Drug-and-Medication-Information>, citing Gerhartz, W. (exec ed.). Ullmann's Encyclopedia of Industrial Chemistry. 5th ed. Vol A1: Deerfield Beach, FL: VCH Publishers, 1985 to Present., p. V2: 523 (1985).

antibiotic resistance with respect to its use in orchards. A statement in the CS proposal by the majority<sup>23</sup> reveals a misunderstanding that is out of sync with the science. Despite the scientific findings to the contrary, the majority concluded: “There is no evidence that applications of antibiotics to orchards during bloom contributes to antibiotic resistance in human pathogens. Human pathogens have not been found in orchards and would have to be present for the resistance genes to transfer.”

The minority position described the current scientific understanding of the spread of resistance through selection and horizontal gene transfer:

Current science shows that environmental exposure to antibiotic use in the environment is the major cause of development and spread of antibiotic resistance in human pathogens.<sup>24</sup> The spread of antibiotic resistance does not require contact between the antibiotic and human pathogens because the major means of spreading antibiotic resistance is through the transfer of genes between different bacteria.<sup>25</sup> Uses resulting in low residues (subtherapeutic or subinhibitory levels) can create a high health risk.<sup>26</sup> [R]esistance is evident and expected to grow if urgent use precaution is not exercised.<sup>27</sup>

Use of antibiotics in fruit is considered to be a serious enough matter by the Infectious Diseases Society of America that it commented to EPA on an emergency exemption request for gentamicin use for fire blight, “There is ample reason to be concerned about adverse human health effects from gentamicin use in plant agriculture...[A] risk assessment model should be developed to understand the potential for adverse health consequences in humans. The FDA has used this approach in guidance to industry for microbial safety of antimicrobial use in food animals, and an equally rigorous approach is needed for use in plant agriculture.”<sup>28</sup> And,

Granting the Michigan Department of Agriculture’s exemption request would set an unfortunate and dangerous precedent during an era when we cannot afford to lose yet another therapeutic option for treating serious infections. IDSA is extremely

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<sup>23</sup> The proposal contained presentations by the majority (favoring extending the use) and the minority (favoring immediate discontinuance of use.)

<sup>24</sup> American Academy of Microbiology, 2009. Antibiotic Resistance: An Ecological Perspective on an Old Problem. <http://academy.asm.org/images/stories/documents/antibioticresistance.pdf> (pp.1-5, 10.)

<sup>25</sup> American Academy of Microbiology, 2009. (p.8.)

<sup>26</sup> <http://www.fda.gov/downloads/AnimalVeterinary/GuidanceComplianceEnforcement/GuidanceforIndustry/UCM052519.pdf>. See Table 6, p.22.

<sup>27</sup> 2011 Tetracycline TR lines 549-551: “There is a high probability that oxytetracycline resistant bacteria are present in the environment as a consequence of pesticidal use of oxytetracycline which may have negative health consequences for humans (EPA, 2006).” American Academy of Microbiology, 2009. p.2: “Controlling antibiotic resistant bacteria and subsequent infections more efficiently necessitates the prudent and responsible use of antibiotics. It is mandatory to prevent the needless use of antibiotics...”

<sup>28</sup> IDSA, April 27, 2006. Letter from Martin J. Blaser, president, to EPA. Re: Comments on Docket ID Number EPA-HQ-OPP-2006-0234 (Receipt of Application for Emergency Exemption: Gentamicin). Pp. 1-2.

concerned about the possibility that the direct or indirect effects of gentamicin use in plant agriculture may seriously compromise gentamicin's effectiveness in the treatment of human bacterial infections. We support further research in this area, but **we strongly oppose even one-time use of gentamicin in plant agriculture until the microbial safety has been thoroughly evaluated.**<sup>4</sup> (Emphasis added.)

At the Spring 2013 NOSB meeting, Glenn Morris, M.D.,<sup>29</sup> professor of infectious diseases in the University of Florida College of Medicine, further refuted the statement that "Human pathogens have not been found in orchards and would have to be present for the resistance genes to transfer," saying (emphasis added):

I can say that the approach that we have started to take increasingly, given the significant problems we're encountering in human medicine, is we need to try to limit or eliminate use in all instances, because all of this—and again, even though, you know, what difference does it make? Well, there is a remote possibility that one could get selection of new tetracycline resistance gene tomorrow, when you spray, and that that could then move into human populations with devastating impact. Again, it's a very, very rare event. But what's fascinating is that when you work with the mathematical models, even very, very rare events can clearly have significant downstream populations because of the potential for amplification once they get into the human intestinal flora. And again, that's our concern.

**It's not movement into a human pathogen, but movement into the overall ecology of your intestinal flora, where it may not even be detectable until you are in a setting where suddenly antibiotics are administered, and where suddenly you become very susceptible to further infection.**

As explained in the TR, "Could kasugamycin orchard sprays lead to bacteria that harbor antibiotic resistance genes? Such is the case with other antibiotics. Spraying orchards with streptomycin and tetracycline can lead to resistant bacteria, such as *Pantoea agglomerans*. These bacteria can release transposons and plasmids that can confer resistance to environmental pathogens such as *Erwinia amylovora*. The same genetic elements that cause resistance in plant pathogens can cause resistance in human pathogens."<sup>30</sup> The TR also describes potential cross-resistance between kasugamycin and other antibiotics.<sup>31</sup> Furthermore, the TR documents the fact that kasugamycin use has led to resistance in epiphytic bacteria, plant pathogens, and river water microcosms.<sup>32</sup> Resistance in fire blight bacteria has been produced in the laboratory.<sup>33</sup>

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<sup>29</sup> Transcript of April 2013 NOSB meeting, page 700 line 13 through page 702 line 6; page 716 line 17 through page 717 line 21.

<sup>30</sup> Kasugamycin TR, lines 774-778.

<sup>31</sup> Kasugamycin TR, lines 849-871.

<sup>32</sup> Kasugamycin TR, lines 554-563; 571-586; 602-605.

<sup>33</sup> Kasugamycin TR, lines 607-615.

## **Kasugamycin disrupts ecological balance in the orchard and surface water.**

The TR points out that EPA's Chronic Risk Quotient is exceeded for several classes of animals who may be found in orchards—mammals of all size who eat short grass; 15g mammals who eat short grass, broadleaf plants, and insects; 35g mammals who eat broadleaf plants and insects. The TR also says, "Risks are exceeded with small mammals. Presumably, there is some risk for larger animals, as the Kasumin label states, 'animal grazing in treated areas is prohibited.'"<sup>34</sup>

Kasugamycin is also phytotoxic to apples and pears.<sup>35</sup>

Although kasugamycin is generally regarded as nontoxic to microorganisms, Huang et al. demonstrated that it inhibits the growth of some aquatic bacteria but also stimulated the growth of other resistant bacteria.<sup>36</sup> The TR documents impacts on plants as well, which could shift the ecological balance of algae and other plants.<sup>37</sup>

## **Kasugamycin use is incompatible with a system of organic and sustainable agriculture.**

The use of antibiotics in organic agriculture is contrary to consumer expectations.<sup>38</sup> It is inconsistent with practices in much of the rest of the world. Reliance on antibiotics is not sustainable because pathogens will develop resistance.

## **Kasugamycin use is not necessary.**

Organic apple and pear production has not ceased since the use of streptomycin and tetracycline was prohibited.<sup>39</sup> The TR describes alternative control strategies. "Biological control can be just as effective as sprays of kasugamycin." "The major mechanism of fire blight biological control is competitive exclusion." "Low rates of copper can be used for shoot blight control during the summer." "Some success has been seen with sprays, trunk paints and injections of materials that cause systemic acquired resistance (SAR)." "The alternative to

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<sup>34</sup> Kasugamycin TR, lines 621-627.

<sup>35</sup> Kasugamycin TR, line 565.

<sup>36</sup> Huang, C.Y., Ho, C.H., Lin, C.J. and Lo, C.C., 2010. Exposure effect of fungicide kasugamycin on bacterial community in natural river sediment. *Journal of Environmental Science and Health Part B*, 45(5), pp.485-491.

<sup>37</sup> Kasugamycin TR, lines 664-670.

<sup>38</sup> As reported in "Organic pome and cherry production and marketing issues: Past, present and future," and presented to IFOAM, "Over the last ten years, the Hartman Group (Bellevue, Washington, USA) has studied changes in consumer attitudes, backgrounds, and buying characteristics related to the organic market. The Hartman Group surveyed about two thousand household consumers across four regions of the USA. They found that the 'traditional' properties suggested by 'organic' were no longer the same properties held by the new organic consumer. The survey indicated that traditional properties such as 'locally-grown,' Fair Trade, 'tastes better,' and sustainable production ranked at the bottom. The new organic consumers made it clear that they want, plain and simple, a product centered around the 'absence of all health concerns,' and the absence of pesticides, growth hormones, GMO's, antibiotics, and BSE."

<sup>39</sup> <https://www.ers.usda.gov/data-products/fruit-and-tree-nuts-data/fruit-and-tree-nuts-yearbook-tables/>.



kasugamycin is an integrated organic program that attacks fire blight at every point in its life cycle.” A great deal more detail is contained in the responses to Questions 11 and 12 in the TR.<sup>40</sup>

**Conclusion: We said “No” to antibiotics in organic fruit, and now we must show that we mean it.**

As we struggle to get free from the coronavirus (SARS-CoV-2) pandemic, we should be especially cognizant of the danger of a “looming potential pandemic” resulting from a “rise in multidrug-resistant bacterial infections that are undetected, underdiagnosed, and increasingly untreatable, [which] threatens the health of people in the USA and globally.”<sup>41</sup> Kasugamycin does not meet any of the OFPA criteria for the National List—it poses health and environmental dangers, is not necessary, and is incompatible with organic practices.

Thank you for your consideration of these comments.

Sincerely,



Terry Shistar, Ph.D.  
Board of Directors

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<sup>40</sup> Kasugamycin TR, lines 877-1112.

<sup>41</sup> Strathdee, S.A., Davies, S.C. and Marcelin, J.R., 2020. Confronting antimicrobial resistance beyond the COVID-19 pandemic and the 2020 US election. *The Lancet*, 396(10257), pp.1050-1053.  
<https://www.thelancet.com/action/showPdf?pii=S0140-6736%2820%2932063-8>.