Understanding arsenic dynamics in agronomic systems to predict and prevent uptake by crop plants

Punshon, T., Jackson, B. P., Meharg, A. A., Warczack, T., Scheckel, K., & Guerinot, M. L. (2017). Understanding arsenic dynamics in agronomic systems to predict and prevent uptake by crop plants. DOI: 10.1016/j.scitotenv.2016.12.111

Published in:
The Science of the total environment

Document Version:
Peer reviewed version

Elsevier Editorial System(tm) for Science of the Total Environment
Manuscript Draft

Manuscript Number: STOTEN-D-16-04444R1

Title: Understanding arsenic dynamics in agronomic systems to predict and prevent uptake by crop plants

Article Type: VSI: Arsenic Pathways to Food

Keywords: arsenic, sources, soil, agriculture, plants, mitigation

Corresponding Author: Dr. Tracy Punshon,

Corresponding Author's Institution: Dartmouth College

First Author: Tracy Punshon

Order of Authors: Tracy Punshon; Brian P Jackson; Andrew A Meharg; Todd M Warczack; Kirk Scheckel; Mary Lou Guerinot

Abstract: This review is on arsenic in agronomic systems, and covers processes that influence the entry of arsenic into the human food supply. The scope is from sources of arsenic (natural and anthropogenic) in soils, biogeochemical and rhizosphere processes that control arsenic speciation and availability, through to mechanisms of uptake by crop plants and potential mitigation strategies. This review makes a case for taking steps to prevent or limit crop uptake of arsenic, wherever possible, and to work toward a long-term solution to the presence of arsenic in agronomic systems. The past two decades have seen important advances in our understanding of how biogeochemical and physiological processes influence human exposure to soil arsenic, and this must now prompt an informed reconsideration and unification of regulations to protect the quality of agricultural and residential soils

Response to Reviewers: Response to Reviewers

We thank both reviewers for taking the time to give our paper a rigorous and thoughtful peer review, and for ultimately improving the quality of the writing and the information. We include our responses to their comments below.

Reviewer 1 Comment 1 (R1C1): Page 3 L15-18: This sentence is a bit misleading. Importantly, As uptake by rice readily occurs in non-contaminated soils because naturally present As is liberated and plant available under reducing soil conditions. The sentence as written suggests that As uptake into rice is due to contamination.

Response to R1C1: We agree; this is an important point. To make it clearer that arsenic contamination of soil is not necessarily a pre-requisite for its entry into food crops, we have added after L15-18: “The presence of elevated concentrations of arsenic in the soil is not a pre-requisite in dietary arsenic exposure as seen in the effective accumulation of arsenic by rice grown in uncontaminated soils.”

R1C2: In addition, I would argue that "in many cases" the route of As exposure via food is not more significant than drinking water. In the US, As ingestion via food is a primary route of exposure because most drinking water in the US has low As. However, in other more populated regions (e.g. S and SE Asia) drinking water is the primary route of exposure and ingestion via food is secondary.

Response to R1C2: We agree: this generalization is a US-centric view. In place of “In many cases”, we have specified that “When drinking-water arsenic concentrations are low, dietary arsenic can be a significant exposure route.”

R1C3: Page 4 L6 and L8 and elsewhere: Is the use of in text URLs suitable for STODEN? Please check. I suggest to cite them in the references instead.

Response to R1C3: Agreed. We have replaced full URLs with citations and moved them to the bibliography.

R1C4: Page 5 L8: It is not clear what the 2.1 mg/kg level is for in Florida. Is that agricultural soils?

Response to R1C4: We have added the missing information. The sentence now reads, “for instance, New Jersey has a cleanup criterion of 20 mg/kg and Florida has a cleanup target of 2.1 mg/kg for residential and 12 mg/kg for industrial sites.”

R1C5:Page 5 L22-23: This is generally true up to a point, but after a threshold is reached As levels in plants may decrease with increasing As in soil or water because As begins to impose plant toxicity, which affects uptake of solutes including As. See, e.g.

⇒ Panaullah, G. M.; Alam, T.; Hossain, M. B.; Loeppert, R. H.; Lauren, J. G.; Meisner, C. A.; Ahmed, Z. U.; Duxbury, J. M., Arsenic toxicity to rice (Oryza sativa L.) in Bangladesh. Plant and Soil 2009, 317, (1-2), 31-39.

⇒ Syu, C. H.; Huang, C. C.; Jiang, P. Y.; Lee, C. H.; Lee, D. Y., Arsenic accumulation and speciation in rice grains influenced by arsenic phytotoxicity and rice genotypes grown in arsenic-elevated paddy soils. Journal of Hazardous Materials 2015, 286, 179-186.

Response to R1C5: We have changed this statement to read, “Below toxic concentrations, the higher the total soil arsenic concentration (the sum of all arsenic species, regardless of bioavailability) the higher the crop uptake of arsenic, including….” and have added the Panaulla and Syu references suggested by Reviewer 1.

R1C6: Page 6 L13: Suggest to change to "most soils range < 7.5 - 20 mg/kg arsenic" for clarity

Response to R1C6: Done.

R1C7: Page 11 L11 and elsewhere: Are edible plant tissue concentrations reported in mg/kg on a fresh weight or dry weight basis? This should be clearly stated to avoid confusion. I assume dry weight, but many of the market basket surveys (especially the FDA total diet study) report on fresh weight. Plants are generally 90% water, so this detail is important when comparing concentrations.

Response to R1C7: Done. This has been stated clearly at first mention.

R1C8: Page 12 L3: There is a hyperlink for "130 million tons" which should instead be removed and referenced.

Response to R1C8: Done.

R1C9: Page 12 L7: change "since" to "because"

Response to R1C9: Done

R1C10: Page 13 L13: Please define DMA and MMA. Not all STODEN readers will know what those compounds are.

Response to R1C10: Done

R1C11: Page 14 L8-15: It should be considered that the microbial communities also differ, and that microbes are known to methylate inorganic As as a detoxification mechanism. DMA and inorganic As differ in their in planta mobility, which is likely a contributing factor in enhanced total As (as DMA) in US vs Bangladeshi rice.

Response to R1C11: To our knowledge there are no comparative studies on the soil microbial communities between (for instance) the USA and Bangladesh that would support this statement, although it is a well-informed hypothesis. We are wary – particularly on what seems to have been a contentious issue – of hypothesizing about why USA rice contains more arsenic, or why the arsenic speciation differs. We have stated however, in response to this comment, what factors may be involved and what the uncertainties are, which will hopefully embrace the comment of Reviewer 1. We have also referenced the statement that plants themselves cannot methylate arsenic. This paragraph now reads:
"Former pesticide application has been suggested be a factor in the presence of higher levels of total arsenic found in rice grown in the south-central regions of the USA65, 66 compared to other areas of the USA and to other countries, such as Bangladesh67. Evidence on varietal differences in arsenic uptake, speciation and distribution within rice grain (See also Section 6) strongly suggest that soil arsenic concentration is not the sole, nor particularly the main driver of this phenomenon. Other factors likely to be influential include the differences in the soil microbial community composition between geographical regions that affect arsenic methylation, which – considering that the ability to methylate arsenic has not been found in plants – may be another driving factor."

R1C12: Page 15 L1-3: It is not clear if the Florida study was from the reference 68? If not, please provide the reference.

Response to R1C12: Done.

R1C13: Page 17 L16: silicate/silicic acid should also be mentioned because it competitively desorbs arsenite. See, eg. Luxton TP, Tadanier CJ, Eick MJ. Mobilization of arsenite by competitive interaction with silicic acid. Soil Sci Soc Am J. 2006;70:204-214.

Response to R1C13: Done.

R1C14: Page 19 L5: What is the reference for the "animal waste products" part of the sentence?

Response to R1C14: Citation has been added.

R1C15: Page 20 L17-19: This sentence is a bit misleading. All plants tend to acidify the rhizosphere due to cation uptake and charge balance by releasing protons to the rhizosphere. In other words, iron uptake by non-grasses is not the only mechanism that results in an acidified pH; those processes (e.g. cation uptake) are universal in all higher plants.

Response to R1C15: Sentence now reads "Rhizosphere acidification occurs in all plant species during cation uptake and charge balance, when protons are released into the rhizosphere".

R1C16: Page 20 L22: for the non-expert, I suggest to explicitly state why high OC in the rhizosphere enhances As solubility (i.e. drives microbially-mediated reductive dissolution).

Response to R1C16: We have added "by stimulating microbially-mediated reductive dissolution of soil minerals"

R1C17: Page 21 L4: It should be "anaerobic" not "aerobic" when discussing submerged rice plants.

Response to R1C17: Done.

R1C18: Page 26 L12-L14. Clear evidence has shown that LSi 1 and 2 transporters are responsible for arsenite uptake into rice roots. These transporters are located on the two casparian strips on the exodermis and the endodermis. Note that no transporter is needed to move the neutral arsenous acid (i.e. arsenite) molecule to the casparian strip, as this can be done apoplastically.

Response to R1C18: Thank you. This statement has been deleted, and the statement "In rice, Low Silicon 1 (OsLsi1) and OsLsi2 are silicic acid transporters and arsenite, MMAV, and DMAV are among their unintended targets148, 149." has been moved up and the paragraph edited accordingly.

R1C19: Page 27 L17-18: The way it is written, it sounds like DMA and MMA start to flow backward "reversed"…I believe what was intended from this sentence is something like "While the inorganic forms are more readily taken up from soil than organic forms, the organic forms are more readily transported to shoots because…".

Response to R1C19: This statement now reads, "Despite having a lower affinity for transporters into the plant than inorganic forms, organic arsenic species are more efficiently transported toward the shoot than inorganic forms."

R1C20: Also, there was no earlier mention of the relative differences between inorganic and organic As for uptake. Consider revising.
Response to R1C20: Statements about the relative differences in uptake of arsenic species are included in two statements in Section 6.2: "In magnitude, plants take up arsenicals from the soil in the order arsenite > arsenate > DMA > MMA" and "The arsenic uptake specificity of OsLsi1 is arsenite >> MMA > DMA".

R1C21: Page 31 L7-19: It should also be mentioned that increased exogenous Si can down-regulated the Lsi1 transporter, which further decreases the potential for As uptake.

Response to R1C21: Added. Thank you.

R1C22: Page 31 L18-19. Suggest to change to "…practice for smallholder farmers), can provide silicon without increasing methane production and decreases either total or inorganic As in grain."

Response to R1C22: Added.

Reviewer 2:
Reviewer 2 Comment 1 (R2C1): As the authors discuss the regulated limits of arsenic in rice, I would have thought a section relating these exposures to arsenic to health impacts was needed. Some indication from the scientific literature on the impacts that arsenic rich water and food has on human health is needed, especially in relation to the concentrations given as the regulated limits.

Response to R2C1: This is a great comment. The reviewer refers to exposure epidemiological studies of arsenic, which is a huge area of science that is outside the scope of this synthesis paper. This topic is the subject of another excellent article submitted to this special issue. We have added a statement referring the reader to that article at the end of the first paragraph of the introduction:
"An in-depth review of the current findings on the relationship between dietary arsenic exposure and human health is provided by Davis et al. (this issue).."

R2C2: Page 4 line 2. "but no safe level of arsenic has been found" what do the authors mean by this statement? The next sentence discusses impacts of arsenic in compared to MCLs, but does not go as far as saying that any concentrations are harmful.

Response to R2C2: We agree that the statement "no safe level of arsenic has been found", suggests that any arsenic level causes actual harm, when in fact observations are that any level causes an effect on biological model systems, such as cell lines. We recognize that this is an important distinction. This statement has been deleted. We have clarified the message here and the statement now reads:
"but given that measurable biological effects occur in at levels below the current maximum contaminant level (MCL) for arsenic in drinking water2, these low levels can still translate into significant exposures, particularly in children and presumably in adults who consume a lot of rice."

R2C3: Page 4 line 18. "the majority of other nations", some clarification needed on this statement as the statement above this indicates that the 10ug/l is supported by WHO, EU and Canada. Does that mean that all other nations except those in the EU, Canada and the couple of other mentioned nations have a limit of 50 ug/l?

Response to R2C3: We have named the countries that still adopt a standard of 50 µg/L, and included a citation.

R2C4: Page 5 line 2. Reference needed for this statement.

Response to R2C4: Done.

R2C5: Page 6 line 21. "USGS" I think this is the first use of this abbreviation. Please spell out in full.

Response to R2C5: Done.

R2C6: Page 9 line 20. A reference is needed for this statement.

Response to R2C6: Done.

R2C7: Page 12 line 3. Formatting of "130 million tons"

Response to R2C7: Done.

R2C8: Page 18 line 8. Can the authors expand on the bind of arsenic to soils particles.

Response to R2C8: We have added the following text to expand on this statement:
"Arsenite is less stably bound to aluminum hydroxides and aluminosilicate clay minerals in the soil than arsenate, for which they exhibit a much stronger binding preference30.

R2C9: Page 25. When discussing toxicity it would be good to indicate what this measurement actually is, is it root assays in hydroponics or soil based systems. As hydroponics is more artificial than the natural soil systems.

Response to R2C9: In our opinion, the discussion about arsenic toxicity in plants is clearly concerned with field studies where reduced yield has been a result. We have clarified this statement, and have ensured that comparative toxicity tests from hydroponic-based assays are not cited.

R2C10: Page 26 line 12. The references related to phosphate transporters, are these the transporters known to be involved in arsenic transport or those involved in phosphate transport and possibly arsenate? It is unclear from the way that it is written.

Response to R2C10: It is our opinion that the statement about arsenate being transported via phosphate transporters is very clear here. We state "Arsenate enters root cells through phosphate transporters" and then we have referenced experimental evidence for this in two model plant species.

R2C11: Page 31, section 7.2. This section seems on the small side given all the potential mechanisms that could mitigate arsenic accumulation by the alteration of arsenic chemistry in the soil. This could be expanded further. Additionally, this is very rice focused (actually all of section 7 is). While I understand that rice is the main target for reducing arsenic due to it being a dietary staple and having very high concentrations, you started the review by discussing not just rice but products like apple juice and vegetables and fruits. I think the mitigation strategies to be balanced need to also focus on non-rice plants to.

Response to R2C11: We agree that this section is short and rice-focused. After re-reviewing the current literature, we added missing information on the use of iron-based amendments, and have expanded the section.
We have restricted our discussion to agricultural settings, and to mesocosms or field-scale studies to highlight technologies closer to deployment, for the article's target audience of stakeholders and regulators. Although we are aware of other amendment types (biochar, kaolin and various other organic matter additions) field scale testing of these materials has been minimal. Another issue – which we have added to the discussion – is that development of amendments proceeds without input from or consideration for the smallholder farmers or commercial entities that would use them, and almost always remain prohibitively priced, or without long term supporting data. We have made reference to this in our conclusions.
In addition, the literature is heavily rice-focused, because these agronomic approaches have been developed in response to the Codex Commission's adoption of a maximum level of 0.2 m/kg inorganic arsenic in polished rice, and because it is primarily in anaerobic flooded systems that arsenic uptake by plants is an issue. We have tried to include studies on other cropping systems where available, but these are inevitably fewer than the rice-based studies.

R2C12: You discuss how water management, fertilisers and the plant can be altered for growing on arsenic soils, I think this could be expanded into possible remediation strategies for the soil as well. A lot of the information that you present in the opening sections is about the addition to arsenic to land / soil, a section on mitigation of this and then dealing with high arsenic soils would be good.

Response to R2C12: This is an interesting topic, but we feel an additional section on phytoremediation of highly arsenic contaminated soils is well beyond the scope of this article. Our focus is on the route arsenic takes into the human food supply, from soils not necessarily classified as highly arsenic contaminated, and how this can be managed to reduce dietary arsenic exposure. A section on mitigation of arsenic input into agricultural soil is also not warranted, since much of the arsenic contamination relevant to food production is either natural, legacy (former pesticide application), or the result of efficient plant-based arsenic accumulation mechanisms. We have dealt with some of these aspects in our conclusion, where we state that information on arsenic levels in soils be made more available so the appropriate action can be taken to avoid human exposure.

*Cover Letter

Department of Biological Sciences • Dartmouth College • Hanover • New Hampshire • 03755 • 603 646 1037

August 19 2016

To Whom It May Concern:,

This letter accompanies the submission of our research article, "**Understanding arsenic dynamics in agronomic systems to predict and prevent uptake by crop plants**". This is an original work. It has never been previously published, in whole or in part, nor is it currently under consideration for publication elsewhere. We declare that there are no actual or potential competing interests, and that all authors have read and approved the manuscript and that they accept responsibility for its contents and agree it is ready for submission.

This paper is part of a series of reviews that resulted from the Collaborative on Food Arsenic and Associated Risk and Regulation, which took place at Dartmouth College, on November 2 2015. The workshop was attended by national and international scientists who lead the field on the study of arsenic in the environment and its affects on our health. The aim of the C-FARR workshop were to identify and synthesize information necessary to advance regulation of arsenic as a food supply contaminant.

This particular review is on arsenic in agronomic systems, and covers processes that influence the entry of arsenic into the human food supply. The scope is from sources of arsenic (natural and anthropogenic) in soils, biogeochemical and rhizosphere processes that control arsenic speciation and availability, through to mechanisms of uptake by crop plants and potential mitigation strategies. This review makes a case for taking steps to prevent or limit crop uptake of arsenic, and to work toward a long-term solution to the presence of arsenic in agronomic systems. The past two decades have seen important advances in our understanding of how sources of arsenic to the soil are influenced by biogeochemistry and result in human exposure, which must now prompt an informed reconsideration and unification of regulations that protect the quality of agricultural and residential soils.

Please note that the review is written to be accessible to a wide range of audiences, from the expert to the informed layperson.

I look forward to a constructive review of our work at Science of the Total Environment

Sincerely,

Response to Reviewers

We thank both reviewers for taking the time to give our paper a rigorous and thoughtful peer review, and for ultimately improving the quality of the writing and the information. We include our responses to their comments below.

Reviewer 1 Comment 1 (R1C1): Page 3 L15-18: This sentence is a bit misleading. Importantly, As uptake by rice readily occurs in non-contaminated soils because naturally present As is liberated and plant available under reducing soil conditions. The sentence as written suggests that As uptake into rice is due to contamination.

Response to R1C1: We agree; this is an important point. To make it clearer that arsenic contamination of soil is not necessarily a pre-requisite for its entry into food crops, we have added after L15-18: "The presence of elevated concentrations of arsenic in the soil is not a pre-requisite in dietary arsenic exposure as seen in the ~~effective~~ accumulation of arsenic by rice grown in uncontaminated soils."

R1C2: In addition, I would argue that "in many cases" the route of As exposure via food is not more significant than drinking water. In the US, As ingestion via food is a primary route of exposure because most drinking water in the US has low As. However, in other more populated regions (e.g. S and SE Asia) drinking water is the primary route of exposure and ingestion via food is secondary.

Response to R1C2: We agree: this generalization is a US-centric view. In place of "In many cases", we have specified that "When drinking-water arsenic concentrations are low, dietary arsenic can be a significant exposure route."

R1C3: Page 4 L6 and L8 and elsewhere: Is the use of in text URLs suitable for STODEN? Please check. I suggest to cite them in the references instead.

Response to R1C3: Agreed. We have replaced full URLs with citations and moved them to the bibliography.

R1C4: Page 5 L8: It is not clear what the 2.1 mg/kg level is for in Florida. Is that agricultural soils?

Response to R1C4: We have added the missing information. The sentence now reads, “for instance, New Jersey has a cleanup criterion of 20 mg/kg and Florida has a cleanup target of 2.1 mg/kg for residential and 12 mg/kg for industrial sites.”

R1C5:Page 5 L22-23: This is generally true up to a point, but after a threshold is reached As levels in plants may decrease with increasing As in soil or water because As begins to impose plant toxicity, which affects uptake of solutes including As. See, e.g.

⇒ Panaullah, G. M.; Alam, T.; Hossain, M. B.; Loeppert, R. H.; Lauren, J. G.; Meisner, C. A.; Ahmed, Z. U.; Duxbury, J. M., Arsenic toxicity to rice (Oryza sativa L.) in Bangladesh. Plant and Soil 2009, 317, (1-2), 31-39.

⇒ Syu, C. H.; Huang, C. C.; Jiang, P. Y.; Lee, C. H.; Lee, D. Y., Arsenic accumulation and speciation in rice grains influenced by arsenic phytotoxicity and rice genotypes grown in arsenic-elevated paddy soils. Journal of Hazardous Materials 2015, 286, 179-186.

Response to R1C5: We have changed this statement to read, “Below toxic concentrations, the higher the total soil arsenic concentration (the sum of all arsenic species, regardless of bioavailability) the higher the crop uptake of arsenic, including….” and have added the Panaulla and Syu references suggested by Reviewer 1.

R1C6: Page 6 L13: Suggest to change to "most soils range < 7.5 - 20 mg/kg arsenic" for clarity

Response to R1C6: Done.

R1C7: Page 11 L11 and elsewhere: Are edible plant tissue concentrations reported in mg/kg on a fresh weight or dry weight basis? This should be clearly stated to avoid confusion. I assume dry weight, but many of the market basket surveys (especially the FDA total diet study) report on fresh weight. Plants are generally 90% water, so this detail is important when comparing concentrations.

Response to R1C7: Done. This has been stated clearly at first mention.

R1C8: Page 12 L3: There is a hyperlink for "130 million tons" which should instead be removed and referenced.

Response to R1C8: Done.

R1C9: Page 12 L7: change "since" to "because"

Response to R1C9: Done

R1C10: Page 13 L13: Please define DMA and MMA. Not all STODEN readers will know what those compounds are.

Response to R1C10: Done

R1C11: Page 14 L8-15: It should be considered that the microbial communities also differ, and that microbes are known to methylate inorganic As as a detoxification mechanism. DMA and inorganic As differ in their in planta mobility, which is likely a contributing factor in enhanced total As (as DMA) in US vs Bangladeshi rice.

Response to R1C11: To our knowledge there are no comparative studies on the soil microbial communities between (for instance) the USA and Bangladesh that would support this statement, although it is a well-informed hypothesis. We are wary – particularly on what seems to have been a contentious issue – of hypothesizing about why USA rice contains more arsenic, or why the arsenic speciation differs. We have stated however, in response to this comment, what factors may be involved and what the uncertainties are, which will hopefully embrace the comment of Reviewer 1. We have also referenced the statement that plants themselves cannot methylate arsenic. This paragraph now reads:
“Former pesticide application has been suggested be a factor in the presence of higher levels of total arsenic found in rice grown in the south-central regions of the USA[65, 66] compared to other areas of the USA and to other countries, such as Bangladesh[67]. Evidence on varietal differences in arsenic uptake, speciation and distribution within rice grain (See also Section 6) strongly suggest that soil arsenic concentration is not the sole, nor particularly the main driver of this phenomenon. Other factors likely to be influential

include the differences in the soil microbial community composition between geographical regions that affect arsenic methylation, which – considering that the ability to methylate arsenic has not been found in plants – may be another driving factor.”

R1C12: Page 15 L1-3: It is not clear if the Florida study was from the reference 68? If not, please provide the reference.

Response to R1C12: Done.

R1C13: Page 17 L16: silicate/silicic acid should also be mentioned because it competitively desorbs arsenite. See, eg. Luxton TP, Tadanier CJ, Eick MJ. Mobilization of arsenite by competitive interaction with silicic acid. Soil Sci Soc Am J. 2006;70:204-214.

Response to R1C13: Done.

R1C14: Page 19 L5: What is the reference for the "animal waste products" part of the sentence?

Response to R1C14: Citation has been added.

R1C15: Page 20 L17-19: This sentence is a bit misleading. All plants tend to acidify the rhizosphere due to cation uptake and charge balance by releasing protons to the rhizosphere. In other words, iron uptake by non-grasses is not the only mechanism that results in an acidified pH; those processes (e.g. cation uptake) are universal in all higher plants.

Response to R1C15: Sentence now reads “Rhizosphere acidification occurs in all plant species during cation uptake and charge balance, when protons are released into the rhizosphere”.

R1C16: Page 20 L22: for the non-expert, I suggest to explicitly state why high OC in the rhizosphere enhances As solubility (i.e. drives microbially-mediated reductive dissolution).

Response to R1C16: We have added “by stimulating microbially-mediated reductive dissolution of soil minerals”

R1C17: Page 21 L4: It should be "anaerobic" not "aerobic" when discussing submerged rice plants.

Response to R1C17: Done.

R1C18: Page 26 L12-L14. Clear evidence has shown that LSi 1 and 2 transporters are responsible for arsenite uptake into rice roots. These transporters are located on the two casparian strips on the exodermis and the endodermis. Note that no transporter is needed to move the neutral arsenous acid (i.e. arsenite) molecule to the casparian strip, as this can be done apoplastically.

Response to R1C18: Thank you. This statement has been deleted, and the statement “In rice, Low Silicon 1 (OsLsi1) and OsLsi2 are silicic acid transporters and arsenite, MMA^V, and DMA^V are among their unintended targets[148, 149].” has been moved up and the paragraph edited accordingly.

R1C19: Page 27 L17-18: The way it is written, it sounds like DMA and MMA start to flow backward "reversed"...I believe what was intended from this sentence is something like "While the inorganic forms are more readily taken up from soil than organic forms, the organic forms are more readily transported to shoots because...".

Response to R1C19: This statement now reads, “Despite having a lower affinity for transporters into the plant than inorganic forms, organic arsenic species are more efficiently transported toward the shoot than inorganic forms.”

R1C20: Also, there was no earlier mention of the relative differences between inorganic and organic As for uptake. Consider revising.

Response to R1C20: Statements about the relative differences in uptake of arsenic species are included in two statements in Section 6.2: “In magnitude, plants take up arsenicals from the soil in the order arsenite > arsenate > DMA > MMA” and “The arsenic uptake specificity of OsLsi1 is arsenite >> MMA > DMA”.

R1C21: Page 31 L7-19: It should also be mentioned that increased exogenous Si can down-regulated the Lsi1 transporter, which further decreases the potential for As uptake.

Response to R1C21: Added. Thank you.

R1C22: Page 31 L18-19. Suggest to change to "...practice for smallholder farmers), can provide silicon without increasing methane production and decreases either total or inorganic As in grain."

Response to R1C22: Added.

Reviewer 2:

Reviewer 2 Comment 1 (R2C1): As the authors discuss the regulated limits of arsenic in rice, I would have thought a section relating these exposures to arsenic to health impacts was needed. Some indication from the scientific literature on the impacts that arsenic rich water and food has on human health is needed, especially in relation to the concentrations given as the regulated limits.

Response to R2C1: This is a great comment. The reviewer refers to exposure epidemiological studies of arsenic, which is a huge area of science that is outside the scope of this synthesis paper. This topic is the subject of another excellent article submitted to this special issue. We have added a statement referring the reader to that article at the end of the first paragraph of the introduction:
“An in-depth review of the current findings on the relationship between dietary arsenic exposure and human health is provided by Davis et al. (this issue)..”

R2C2: Page 4 line 2. "but no safe level of arsenic has been found" what do the authors mean by this statement? The next sentence discusses impacts of arsenic in compared to MCLs, but does not go as far as saying that any concentrations are harmful.

Response to R2C2: We agree that the statement “no safe level of arsenic has been found”, suggests that any arsenic level causes actual harm, when in fact observations are that any level causes an effect on biological model systems, such as cell lines. We recognize that this

is an important distinction. This statement has been deleted. We have clarified the message here and the statement now reads:
“but given that measurable biological effects occur in at levels below the current maximum contaminant level (MCL) for arsenic in drinking water[2], these low levels can still translate into significant exposures, particularly in children and presumably in adults who consume a lot of rice.”

R2C3: Page 4 line 18. "the majority of other nations", some clarification needed on this statement as the statement above this indicates that the 10ug/l is supported by WHO, EU and Canada. Does that mean that all other nations except those in the EU, Canada and the couple of other mentioned nations have a limit of 50 ug/l?

Response to R2C3: We have named the countries that still adopt a standard of 50 µg/L, and included a citation.

R2C4: Page 5 line 2. Reference needed for this statement.

Response to R2C4: Done.

R2C5: Page 6 line 21. "USGS" I think this is the first use of this abbreviation. Please spell out in full.

Response to R2C5: Done.

R2C6: Page 9 line 20. A reference is needed for this statement.

Response to R2C6: Done.

R2C7: Page 12 line 3. Formatting of "130 million tons"

Response to R2C7: Done.

R2C8: Page 18 line 8. Can the authors expand on the bind of arsenic to soils particles.

Response to R2C8: We have added the following text to expand on this statement: “Arsenite is less stably bound to aluminum hydroxides and aluminosilicate clay minerals in the soil than arsenate, for which they exhibit a much stronger binding preference[30].

R2C9: Page 25. When discussing toxicity it would be good to indicate what this measurement actually is, is it root assays in hydroponics or soil based systems. As hydroponics is more artificial than the natural soil systems.

Response to R2C9: In our opinion, the discussion about arsenic toxicity in plants is clearly concerned with field studies where reduced yield has been a result. We have clarified this statement, and have ensured that comparative toxicity tests from hydroponic-based assays are not cited.

R2C10: Page 26 line 12. The references related to phosphate transporters, are these the transporters known to be involved in arsenic transport or those involved in phosphate transport and possibly arsenate? It is unclear from the way that it is written.

Response to R2C10: It is our opinion that the statement about arsenate being transported via phosphate transporters is very clear here. We state “Arsenate enters root cells through phosphate transporters” and then we have referenced experimental evidence for this in two model plant species.

R2C11: Page 31, section 7.2. This section seems on the small side given all the potential mechanisms that could mitigate arsenic accumulation by the alteration of arsenic chemistry in the soil. This could be expanded further. Additionally, this is very rice focused (actually all of section 7 is). While I understand that rice is the main target for reducing arsenic due to it being a dietary staple and having very high concentrations, you started the review by discussing not just rice but products like apple juice and vegetables and fruits. I think the mitigation strategies to be balanced need to also focus on non-rice plants to.

Response to R2C11: We agree that this section is short and rice-focused. After re-reviewing the current literature, we added missing information on the use of iron-based amendments, and have expanded the section.

We have restricted our discussion to agricultural settings, and to mesocosms or field-scale studies to highlight technologies closer to deployment, for the article's target audience of stakeholders and regulators. Although we are aware of other amendment types (biochar, kaolin and various other organic matter additions) field scale testing of these materials has been minimal. Another issue – which we have added to the discussion – is that development of amendments proceeds without input from or consideration for the smallholder farmers or commercial entities that would use them, and almost always remain prohibitively priced, or without long term supporting data. We have made reference to this in our conclusions.
In addition, the literature is heavily rice-focused, because these agronomic approaches have been developed in response to the Codex Commission's adoption of a maximum level of 0.2 m/kg inorganic arsenic in polished rice, and because it is primarily in anaerobic flooded systems that arsenic uptake by plants is an issue. We have tried to include studies on other cropping systems where available, but these are inevitably fewer than the rice-based studies.

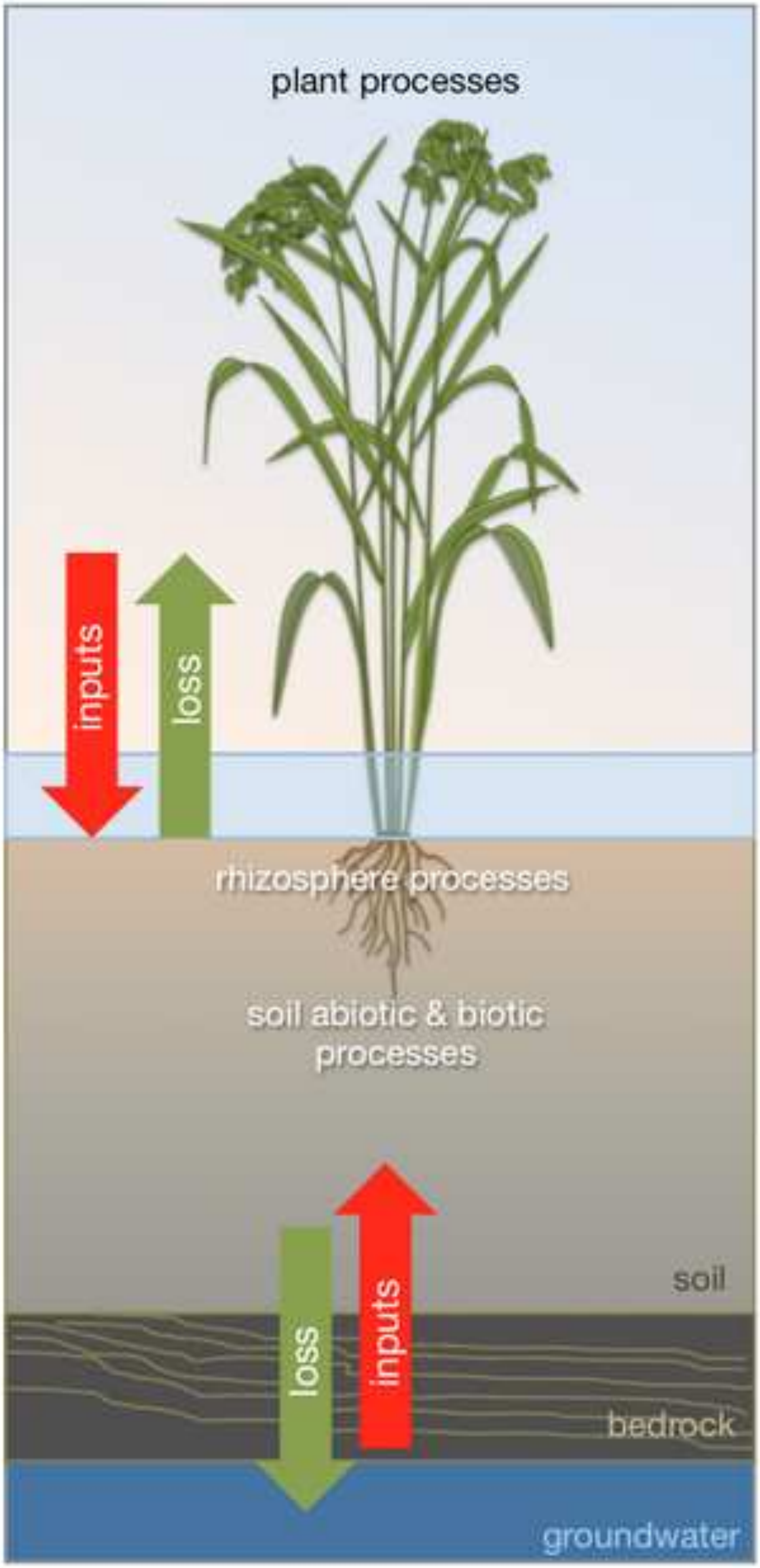
R2C12: You discuss how water management, fertilisers and the plant can be altered for growing on arsenic soils, I think this could be expanded into possible remediation strategies for the soil as well. A lot of the information that you present in the opening sections is about the addition to arsenic to land / soil, a section on mitigation of this and then dealing with high arsenic soils would be good.

Response to R2C12: This is an interesting topic, but we feel an additional section on phytoremediation of highly arsenic contaminated soils is well beyond the scope of this article. Our focus is on the route arsenic takes into the human food supply, from soils not necessarily classified as highly arsenic contaminated, and how this can be managed to reduce dietary arsenic exposure. A section on mitigation of arsenic input into agricultural soil is also not warranted, since much of the arsenic contamination relevant to food production is either natural, legacy (former pesticide application), or the result of efficient plant-based arsenic accumulation mechanisms. We have dealt with some of these aspects in our conclusion, where we state that information on arsenic levels in soils be made more available so the appropriate action can be taken to avoid human exposure.

***Graphical Abstract**

C-FARR

Collaborative on Food with Arsenic and associated Risk and Regulation



Highlights

Understanding arsenic dynamics in agronomic systems to predict and prevent uptake by crop plants

- Consumption of staple foods such as rice, apple juice and vegetables grown in contaminated soil is now recognized as a tangible route of human exposure to arsenic
- Arsenic occurs in food because it is present in the soil and water and is taken up by crop plants.
- Understanding the sources of arsenic to crop plants and influence the dynamics of the agronomic arsenic cycle are key to reducing crop uptake of arsenic now, and preventing exposure in future.
- This review considers natural and anthropogenic sources of arsenic to the soil, biogeochemical cycling, rhizosphere processes, plant processes, and mitigation strategies
- There must be strategies in place that protect human health from soil contamination by arsenic.
- This review recommends: mobilizing existing soil data so that it is readily accessible to commercial and private growers; expanding detailed soil monitoring; reconsideration, unification and enforcement of action levels for agricultural soil arsenic based on updated science, community outreach and education about the potential for arsenic in the soil, as necessary steps to protecting valuable soil resources.

Understanding arsenic dynamics in agronomic systems to predict and prevent uptake by crop plants.

Tracy Punshon*[1], Brian P. Jackson[2], Andrew A. Meharg[3], ~~Mary Lou Guerinot[1],~~ Todd Warczack[1], Kirk Scheckel[4]and Mary Lou Guerinot[1].~~Kirk Scheckel[4]~~

*Corresponding author:

[1]Dartmouth College, Department of Biology, 78 College Street, Hanover, New Hampshire, 03755, USA. tracy.punshon@dartmouth.edu, mary.lou.guerinot@dartmouth.edu, Todd.M.Warczack.GR@dartmouth.edu

[2]Dartmouth College, Department of Earth Sciences, Hanover, New Hampshire, 03755, USA. bpj@dartmouth.edu

[2]Institute for Global Food Security, Queen's University Belfast, Belfast BT9 5HN, United Kingdom. aa.meharg@qub.ac.uk

[4]USEPA Office of Research and Development, National Risk Management Laboratory, 26 West Martin Luther King Drive, Cincinnati, OH 45224. scheckel.kirk@epa.gov

Keywords: arsenic, sources, soil, agriculture, plants, mitigation

Abstract

This review is on arsenic in agronomic systems, and covers processes that influence the entry of arsenic into the human food supply. The scope is from sources of arsenic (natural and anthropogenic) in soils, biogeochemical and rhizosphere processes that control arsenic speciation and availability, through to mechanisms of uptake by crop plants and potential mitigation strategies. This review makes a case for taking steps to prevent or limit crop uptake of arsenic, wherever possible, and to work toward a long-term solution to the presence of arsenic in agronomic systems. The past two decades have seen important advances in our understanding of how ~~sources of arsenic to the soil are influenced by~~ biogeochemical~~stry~~ and physiological processes influence ~~result in~~ human exposure to soil arsenic, ~~which~~ and this must now prompt an informed reconsideration and unification of regulations ~~that~~ to protect the quality of agricultural and residential soils.

1. Introduction

Consumption of staple foods such as rice, beverages such as apple juice, or vegetables grown in historically arsenic-contaminated soils are~~is~~ now recognized as ~~a~~ tangible routes of arsenic exposure~~-~~. The presence of elevated concentrations of arsenic in the soil is not a pre-requisite for dietary arsenic exposure; seen in the accumulation of arsenic by rice grown in uncontaminated soils[1]. ~~that,~~ When drinking-water arsenic concentrations are low, dietary arsenic can be a significant exposure~~in many cases, is more significant than exposure from drinking water~~[2]. Understanding the sources of arsenic to crop plants and the factors that influence them is key to reducing human exposure now and preventing exposure in future. In addition to the abundant natural sources of arsenic, there are a large number of industrial and agricultural sources of arsenic to the soil; from mining wastes, coal fly ash, glass manufacturing, pesticide application, wastewater sludge, pharmaceutical waste, livestock dips, smelting activities to phosphate fertilizers. Plant uptake of arsenic was previously assumed to be too low to merit setting limits for arsenic in food crops, but given that ~~no safe level of arsenic has been found;~~ measurable biological effects occur in at arsenic levels below the current maximum contaminant level (MCL) for ~~arsenic in~~ drinking water[3], these low levels can still translate into significant exposures, particularly in children[4] and presumably in adults who consume a lot of rice. In response, the World Health Organization (WHO) ~~who~~ set an advisory MCL for inorganic arsenic in white (polished) rice of 0.2 mg/kg [5]~~(http://www.who.int/ipcs/assessment/public_health/arsenic/en/)~~ along with the limit of 10 µg/L in water, and the European Union set similar standards that included a lower MCL (0.1 mg/kg) for rice-containing baby foods[6] ~~(http://eur-lex.europa.eu/legal-contect/EN/TXT/?uri=urisrv%3AOJ.L_2015.161.01.0014.01.ENG)~~. Currently, dietary arsenic exposure is suspected to play a role in cardiovascular disease in adults[7], and to disrupt the glucocorticoid system (involved in learning and memory) to those exposed *in utero*[8]. An in depth review of the current findings on the relationship between dietary arsenic exposure and human health is provided by Davis et al. (this issue).

In the United States, regulations on arsenic are distributed to several agencies. The Environmental Protection Agency (EPA) developed the MCL for arsenic in drinking water (10 μg/L) in 2006; a level supported by the World Health Organization, Canada and the European Union. In the state of New Jersey (USA) the limit is 5 μg/L, and in Australia, 7 μg/L. ~~The majority of~~Many other nations still adopt a level of 50 μg/L (Bahrain, Bangladesh, Bolivia, China, Egypt, India, Indonesia, Oman, Philippines, Saudi Arabia, Sri Lanka, Vietnam, Zimbabwe)[9], with the exception of Mexico (35 μg/L). In the USA, The Food and Drug Administration (FDA) is responsible for setting action levels for arsenic in food, which includes apple and pear juice at 10 μg/L, in line with EPA's drinking water MCL. In Canada, the Canadian Food Inspection Agency issued alerts on excessive arsenic in rice and pear products in 2014. Consistent with the European Commission's limit for arsenic in rice used in food production for infants and young children, the FDA is proposing an action level of 0.1 mg/kg for inorganic arsenic in infant rice cereal[10]. Foods in Australia and New Zealand may not contain more than 1 mg/kg dry mass of arsenic, and salt for food use must not contain more than 0.5 mg/kg. Japan has a limit of 15 mg/kg of arsenic in paddy soils ~~(http://www.env.go.jp/en/water/soil/sp.html~~[11]~~)~~. ~~Likewise~~Likewise, Thailand has an agricultural arsenic soil quality standard of 3.9 mg/kg. Within the USA, states differ widely in their action levels for arsenic in soil, for instance New Jersey has a cleanup criterion of 20 mg/kg and Florida has a cleanup target level of 2.1 mg/kg and 12 mg/kg for industrial sites[12].

Arsenic occurs in food because it is present in soil and water and is taken up by plants. This review article brings together the latest scientific information on arsenic in agronomic systems, describing its sources in soils and the processes that influence the uptake of arsenic by crop plants. The intention of this review is to prompt a reconsideration and unification of government regulations on action levels for arsenic in agricultural soil; raise awareness of how both former and ongoing inputs of arsenic to soil can result in food contamination and impacts to human health and finally, to indicate the way forward for mitigation strategies that safeguard valuable soil resources.

2. Natural sources of arsenic in soil

Below toxic concentrations, t~~T~~he higher the total soil arsenic concentration (the sum of all arsenic species, regardless of bioavailability) the higher the crop uptake of arsenic.~~,~~ ~~including~~ This is true of anaerobic cultivation systems such as rice[13-15], aerobic horticultural systems[16] as well as conventional (aerobic) agriculture[15]. The global average total soil arsenic concentration is 5 mg/kg, (equivalent to parts per million), but there is large variation between and within geographical regions[17]. Where soils have formed on arsenic-rich bedrock, or downstream of these bedrocks, very high concentrations of natural arsenic can result. Concentrations of up to 4000 mg/kg arsenic have been measured in soils from the arsenopyrite belt (iron arsenic sulphide, FeAsS) in Styria, Austria[18], for instance. There are approximately 568 known minerals that contain arsenic as a critical component[19]. Arsenic is present in many rock-forming minerals because it can chemically substitute for phosphorus (V), silicate (IV), aluminum (III), iron (III) and titanium (IV) in mineral structures. Global mapping data of total arsenic concentrations in topsoil is not available, although~~.~~ l~~L~~arge-scale regional maps are available for soil arsenic concentrations in Europe[20] and the USA [21]. European data predicts that most soils ~~are ~~range ~~below ~~< 7.5 - 20 mg/kg arsenic, with a median of 6 mg/kg[20]. This prediction comes from block regression-kriging; a spatial prediction technique based on regressing soil arsenic concentrations against auxiliary variables, and is useful because it uses a particularly high resolution (block size of 5 km^2). On a continental scale, large zones of soils with approximately 30 mg/kg arsenic have been found in southern France, the ~~northeastern~~north-eastern Iberian Peninsula and south-west England, with the two latter being zones of extensive natural mineralization associated with base and precious metal mining activities. The United State Geological Survey (USGS) soil sampling of the contiguous USA reports a mean USA soil arsenic concentration of approximately 5 mg/kg with 5 and 95 percentile values of approximately 1.3 and 13 mg/kg respectively[22]. Large regional patterns are apparent in the data, for example the soils of New Hampshire have soil arsenic concentrations of approximately 10 mg/kg arsenic, and Florida, 3.5 mg/kg. The sampling density goal for the USA surface soils and stream sediments database is 1 per 289 km^2 [23], but is currently at only 1 sample per 1600 km^2. This contrasts with smaller regional surveys such as the recently published Tellus database for Northern Ireland that has a sampling density of 2 km^2 [24] (median total soil arsenic concentration 8.7 mg/kg). At this sampling density, fine-scale data for factors shown to affect soil arsenic, such as bedrock type, altitude and organic matter for instance, can be observed, providing the opportunity to make predictions about arsenic bioavailability and mobility.

Soil or sediment arsenic concentrations are the result of the complex and dynamic interplay between inputs and outputs[25]. Natural sources of arsenic to agronomic catchments are dominated by bedrock weathering (mechanical, chemical and biological) and depositional inputs, with the ultimate sinks at the base of catchments often being a significant distance ~~away~~ from sources[26]. Outputs include leaching into water bodies (vertically and horizontally), soil erosion[25] and biovolatilization[27]. In arid regions surface evaporation of water can lead to arsenic enrichment from the draw up of subsurface water[25] and from waters used in crop irrigation[28]. Mass-balances (accounting for all inputs and outputs for a particular ecosystem) are rarely conducted for arsenic fluxes within catchment areas, but a good example is from a mining-impacted catchment area[29], where chemical weathering, followed by mechanical weathering dominated arsenic inputs, which were primarily from arsenopyrite. Similarly, in a gold-mining region, weathering contributed an estimated 95% of the arsenic[30]. In a forested catchment area, where atmospheric arsenic inputs were the dominant source to highly organic soil (soils with more than 10% organic matter), inputs of arsenic via precipitation were ~6 g/ha/y[31], and organic soils were a net source of arsenic, while mineral soils (~~soils with~~ less than 10% organic matter) were a sink. This agrees with depositional inputs of arsenic measured in the UK, which ranged from ~1 to ~10 g arsenic/ha/y[32]. UK regional scale maps show that arsenic deposition is highest at altitude and in the west of the country; the least polluted regions with air masses originating in the Atlantic. This suggests a marine source of arsenic. Depositional maps relate well to soil arsenic maps such as in maps of Northern Ireland and England[33] that show highest arsenic concentrations in peat soils at higher altitude, along with bedrock geological anomalies. Peat soils at higher altitude are sinks for arsenic, and become sources if the peat is mineralized or eroded. The topic of upland organic soils acting as sinks and sources of arsenic is receiving more research attention[34], and could be important on a regional scale as a source of arsenic to downstream sediments.

In large catchment areas of continental importance, such as the deltas that form to the south and east of the Himalayas, plate tectonic-derived mechanical weathering is thought to be the most important source of arsenic. One theory is that the mechanical weathering caused by Pleistocene tectonic uplift in the Himalayas is the key to understanding why arsenic is so elevated in Holocene aquifers, such as those of SE Asia, and in the glacial tills of Europe and North America[26]. Mechanical weathering of bedrock exposes previously inaccessible mineral surfaces, and the finer grinding leads to enhanced surface areas for chemical and microbial weathering to take place, causing greater solubilisation of arsenic[25, 26, 35]. Chemical and microbial weathering can take place at or near the source, or in sediment sinks.~~;~~ F~~f~~or ~~instance~~instance, bacteria isolated from Bay of Bengal aquifers can mobilize arsenic from apatite[35] (See Section 5). Invariably, the arsenic loadings into soil will be dependent on arsenic in the bedrock, and the extent of the weathering of that bedrock-derived material along the route from source to sink. Soils with basalt bedrock had the lowest median arsenic content, while those with psammite, semipelite, and lithic arsenite bedrocks had the highest. Interpretation of such fine-scale mapping can ultimately lead to predictions of soil arsenic concentrations where detailed maps are not available. Combined with an understanding of soil chemistry, this will enhance the ability to predict elevated concentrations of arsenic in crops[36].

3. Anthropogenic sources of arsenic to soil

Many anthropogenic activities have increased soil arsenic concentrations above the natural, background levels mentioned in Section 2 above, and they have the potential to increase the arsenic concentration in food. This is especially the case in the USA where the widespread use of arsenic-based herbicides, pesticides and livestock antibiotics throughout the 20th century has ultimately increased the arsenic concentrations of current productive USA agricultural soils[37-39].

3.1. Base and precious metal mining

The dominant mineral source of arsenic is thought to be pyrite (iron sulfide, FeS_2)[40], an economically important ore deposit. High arsenic concentrations are found in many oxide minerals and hydrous metal oxides, either part of their structure or as sorbed and occluded species[41]. Iron oxides accumulate arsenic up to concentrations of several weight percent (1 weight % being equivalent to 10,000 mg/kg), and arsenic tends to bind to iron (III) (hydr)oxides whenever they are present. Arsenic is found predominantly as arsenopyrite but also can occur as orpiment (arsenic trisulphide As_2S_3), realgar (α-As_4S_4) and other arsenic sulfide minerals[42, 43]. Arsenic is a byproduct of most mining operations and is present at high concentrations in the mine waste, and, because arsenic sulfides are particularly prone to oxidation in surface environments, in mining wastewaters[42, 44]. Arsenic can constitute 1% or more of the ore and solid waste, and wastewaters and impacted streams often contain dissolved arsenic concentrations ranging from 0.01 to over 10 mg/L. Because mining and smelting operations are localized, arsenic contamination of soils exists around the mine site with the concentration decreasing with distance from the source. Windblown dispersion of fine particulate material is a particular problem, spreading contamination greater distances from the mine site. This fine material - which is not completely removed by washing[16] - can directly contaminate plant material; especially leafy material with high surface area. This presents a tangible risk to residents and home gardeners in the vicinity of areas with significant surface soil arsenic contamination. A comparison of arsenic concentrations in vegetables grown in SW England (the site of historic mining activities) with those from a pristine site in NW Scotland found a generally good correlation between total plant arsenic and soil arsenic concentrations. Increased arsenic concentrations were measured in produce from SW England where soil arsenic concentrations ranged from 120 – 1130 mg/kg. Arsenic concentrations were high in leafy greens (kale, spinach, lettuce) and some unpeeled vegetables (potatoes, swedes, carrots) were higher than when peeled, which, in both cases, points to contamination from windblown soil particles and soil adhesion to below ground biomass, rather than from root uptake. In this particular study, the majority of arsenic was present as the inorganic form[16]. Similar results were obtained from home gardens near the Iron King Mine Superfund Site in Arizona, USA[45, 46]. Here the tailings had arsenic concentrations of 3,710 mg/kg and residential soil sampled adjacent to the site ranged from 120 – 633 mg/kg. Edible plant tissue concentrations ranged from < 0.01 – 1.96 mg/kg (plant concentrations are expressed as dry weight throughout), and were generally positively correlated with soil arsenic concentrations. Leafy and high surface area vegetables such as lettuce, kale, broccoli and cabbage accumulated higher arsenic concentrations than beans, tomatoes, cucumbers and peppers. Arsenic in mine-affected vineyard soils in Italy ranged from 4 – 283 mg/kg and positive correlations were observed between soil concentrations and arsenic levels in vine leaves and grapes, however, levels in wine were low (< 1.62 µg/L)[47]. In the Hunan province, China, the high levels of inorganic arsenic in rice have been traced to mining activities in the area[48-50].

3.2. Coal combustion for energy

The concentration of arsenic in USA coal ranges from 1 – 71 mg/kg with an average concentration of 24 mg/kg[51]. Fly ash, the major byproduct of the coal combustion process, consists of fine particles that are driven out with the flue gases, and is a major source of arsenic to the wider environment. Coal ash is one of the most abundant of industrial wastes; close to 130 million tons[52] ~~130 million tons~~ of coal fly was generated in the USA in 2014, with 100 million tons estimated from the European Union in 2011[53]. Arsenic concentrates in the fly ash during combustion of coal for energy; the median arsenic concentration in USA fly ash is 71 mg/kg[54]. Fly ash is often sluiced into settling basins, and ~~since~~ because arsenic in fresh ash is quite soluble, wastewater arsenic concentrations can consequently be quite high.

Formatted: Default Paragraph Font

Arsenic can build up in the sediments of coal fly ash settling basins and reach concentrations of over 1000 mg/kg. Catastrophic failures of these setting basins have caused severe environmental problems and contaminated surface waters with arsenic[55]. There is a well-founded concern that arsenic from coal combustion wastes can contaminate soil and enter the food supply. The use of coal fly ash as a soil amendment can lead to elevated arsenic concentrations in crops (as well as boron, selenium and molybdenum), although its lack of soil macronutrients and the potential for arsenic toxicity prevents the sole application of coal fly ash as a soil amendment[56, 57]. Formulating ash/organic waste mixtures that conform to USEPA regulations for total arsenic application and meet soil and plant fertility requirements has been shown to safe and effective for agronomic use[58].

3.3. Pesticides

Perhaps the largest anthropogenic input of arsenic to agricultural soils in the USA is from the agricultural use of arsenic-based pesticides and herbicides for most of the 20th century. Calcium arsenate and lead arsenate were used extensively up to the 1950s, mostly on orchard soils to combat the codling moth. At peak, 132,000 metric tons of each pesticide compound was applied annually between 1930-1940[37]. In addition to apples, inorganic arsenic pesticides were used on a range of crops including essentially all fruit trees, vine berries, sweet potatoes, white potatoes, most vegetables and cotton[37]. Both lead and arsenate have long residence times in soils and high concentrations (often >100 mg/kg) of these two elements have been reported in old orchard soils in Washington[59], North Carolina[60], New Hampshire[61], New Jersey[62] and Virginia[63]. There is some evidence of greater mobility for arsenic (than lead)[61, 64], and retention of both elements depends on soil type and other environmental factors but most of this legacy contaminant remains in the soil[62]. Use of lead arsenate decreased after 1950s and was finally banned in 1988. The organic arsenic compounds dimethylarsinic acid (DMA) and monomethylarsonic acid (MMA) were used as pesticides on cotton and herbicides for golf courses and right-of-ways until they too were withdrawn from use in 2013. High levels of MMA were reported in transient surface waters adjacent to a crop sprayer operation[65]. Legacy soil arsenic contamination resulting from organic arsenical pesticides plays a major role in straighthead disease of rice[66] (See Section 6.1). It may be that arsenical pesticides have leached to groundwater, as has been suggested for the Texas High Plains Aquifer[67], although a study of the Ogallala aquifer in the High Plains in Texas found no evidence of anthropogenic arsenic in the groundwater[68]. Similarly, there was no relationship between groundwater arsenic and past (inorganic) arsenic pesticide usage in a comprehensive study of New Hampshire groundwater sources[69]. About 10%, depending on soil substrate, of monosodium methyl arsenate applied to sandy soils (simulated golf course greens) leached into percolating water.~~ and~~ D~~d~~emethylation and methylation occurred because both inorganic arsenic species and DMA were also detected in the percolating water[70]. As with mining-impacted soils, plants grown on soils that are high in arsenic from arsenical pesticide contamination take up higher levels of arsenic into their edible tissues, observed for example in potatoes[71], carrots[72] and leafy green vegetables[73, 74].

Former pesticide application has been suggested be a factor in the presence of higher levels of total arsenic found in rice grown in the south-central regions of the USA[75, 76] compared to other areas of the USA and to other countries, such as Bangladesh[77]. ~~A survey of arsenic concentrations in whole grain rice products purchased from various countries found that rice grown in the USA and EU contained more total arsenic than rice grown with arsenic-contaminated groundwater irrigation in Bangladesh[67].~~ Evidence on varietal differences in arsenic uptake, speciation and distribution within rice grain (See also Section 6) strongly suggest that soil arsenic concentration is not the sole, nor particularly the main driver of this phenomenon. ~~{Meharg, 2004 #448}{Zavala, 2008 #449}~~ Other factors likely to be influential include the differences in the soil microbial community composition between geographical regions that affect arsenic methylation, considering that plants themselves cannot methylate arsenic[78]. ~~The distribution of arsenic forms found in the grains differed between cultivars, and is discussed in more detail in Section 6.~~

3.4. Wood preservatives

Chromated copper arsenate (CCA) is used as a wood preservative and was extensively used on decking~~,~~ and other residential usages until a voluntary manufacturer withdrawal in 2003. The primary health concern is for young children in direct contact with CCA-treated wood, but localized leaching of arsenic (as well as chromium and copper) also occurs to surrounding soil. Soil arsenic concentrations of 37 – 250 mg/kg have been reported for soils sampled near CCA-treated utility poles (N=12)[79] and mean arsenic concentrations for soils collected below decks and footbridges in Florida, USA was reported to be 28.5 mg/kg compared with a control concentration of 1.3 mg/kg (N= 65)[80]. Arsenic from CCA contaminated soils appears to be more bioavailable than from other anthropogenic sources to soil[81].

3.4. Organic manures

Land application of sewage sludge (biosolids) in the USA is regulated by Environmental Protection Agency Part 503 Biosolids rule; which set the maximum arsenic concentration of the sludge at 75 mg/kg, an annual pollutant-loading rate of 2.0 kilograms arsenic/hectare~~a~~ (kg/ha) and a cumulative pollution-loading rate of 41 kg/ha over the lifetime of applications. Assuming a plow layer of 17 cm, application at the maximum annual rate implies an approximate 1.2 mg/kg maximum increase in soil arsenic, while the cumulative maximum loading rate could increase soil arsenic concentrations by approximately 24 mg/kg over the lifetime of application and assuming no loss from the soil profile. This cumulative loading rate of 24 mg/kg is significant when considered against an average soil arsenic concentration of 5 mg/kg (See Section 2), however, relative to mine-impacted or inorganic arsenic pesticide impacted soils where arsenic concentrations are frequently > 100 mg/kg, it is of lesser concern. Also, sewage sludge is often high in aluminum and iron oxide phases, used in the flocculation process, which are efficient scavengers of inorganic arsenic thus lowering the arsenic bioavailability [82].

Arsenic occurs in animal wastes primarily because of the former use of arsenic antibiotics in poultry and turkey feed; until 2015 four drugs, roxarsone, p-arsanilic acid, carbarsone and nitrosone, were regulated for use, with roxarsone being the most prevalent. As of 2016 all four of these compounds have all been withdrawn from use[83]. All four are organic arsenic compounds with an arsenate functional group attached to a benzene ring, and differ by other substituents on the ring. The compounds are not readily adsorbed or metabolized and so occur at concentrations up to 40 mg/kg in animal manures. This provides three points of entry to the human food chain; directly through arsenic in chicken and turkey meat[38, 39, 84], from plant uptake after land application of manure, and runoff to surface water or groundwater. A number of studies have shown that these organic arsenic compounds can be degraded by both photolytic[85] and microbial[86] processes and that this degradation happens both during composting of stockpiled litter[87, 88] and after land application[89, 90]. Long term application of poultry litter to Upper Coastal Plain soils increased~~s~~ soil arsenic concentrations~~;~~ from 2.7 to 8.4 mg/kg ~~for an Upper Coastal Plain soil~~ after 25 years of application[91]. ~~and~~ S~~s~~imilar increases have been reported for other southern states of the USA[92]. There is evidence to suggest that other soluble constituents of the litter, for example phosphate and dissolved organic carbon compounds, facilitate arsenic solubility and leaching[89, 92, 93].

3.5. Seaweed fertilizers

Seaweeds can contain far higher concentrations of arsenic than crop plants: up to 100 mg/kg (~~See~~ Taylor et al, this issue). In most cases the arsenic is present as arsenosugars, which are of low toxicity to humans (~~See~~ Taylor et al, this issue). However, as in the case of poultry litter, these compounds degrade (ultimately) to inorganic arsenic after land application[94]. Although seaweeds are a ‘niche’ soil amendment, their use agriculture is increasing and has been adopted by many organic farms as a soil fertilizer as well as a feed additive in organic dairy farming[95].

4. Biogeochemical cycling within terrestrial agronomic ecosystems

Arsenic cycles within the soil surface and near-surface environment[96], influenced by mineralogy, abiotic factors such as pH and redox potential (E_H), and biotic factors such as microbially-mediated biomethylation.

4.1. Redox regulation

The most important biogeochemical step in the exposure of humans to arsenic is its release from soils and sediments into pore water; the water contained within soil pores and/or rock[40]. With the exception of extreme pH conditions (<4 or >9), or high concentrations of competing ions (e.g. phosphate, silicic acid or silicate[97]) the release of arsenic from its strong bonds with soil particles depends upon redox potential (E_H); the extent of aeration of the soil[40]. As E_H falls, electron acceptors are depleted and anoxic conditions develop, causing iron oxides and oxyhydroxides to be reduced and dissolve, releasing sorbed arsenic into the soil solution[98] where it can be taken up by plant roots, or leached into groundwater.

Agronomic cropping systems can be divided with respect to arsenic mobilization on the basis of their redox status. Dominant biogeochemical processes influencing aerobic systems, specifically cereals, upland rice, fruit tree orchards, and community gardens, differ from those that dominate in anaerobic systems, predominantly in flooded rice paddies. In aerobic soils, arsenic speciation is predominantly arsenate (arsenic (V)), and is tightly bound to soil particles. Under anaerobic or flooded conditions, arsenic is reduced, and arsenite (arsenic (III)) is the dominant species[40]. Arsenite is less stably bound to aluminum hydroxides and aluminosilicate clay minerals in the soil ~~particles~~ than arsenate, for which they exhibit a much stronger binding preference[40]. With few exceptions (such as under conditions of sulfur release), transition of arsenic speciation from arsenate to arsenite is the most influential factor to arsenic bioavailability; and it is under anaerobic conditions where arsenic becomes an imminent human health concern. Influential biogeochemical processes in aerobic systems are ageing and accumulation of arsenic in soil, and in anaerobic systems reductive dissolution of iron-bearing minerals is the dominant process.

4.2. Biotransformation to methylated and volatile species

Volatile arsenicals are arsenic species with a boiling point below 150˚C; the most volatile of which is arsine gas (AsH_3), followed by monomethylarsine ($MeAsH_2$), dimethylarsine (Me_2AsH_2) and finally completely methylated trimethylarsine (TMA). Volatile arsenic species can be formed either biotically – by fungi, bacteria and algae[27, 99, 100]or abiotically[99]. In natural systems arsines readily react with oxygen to form non-volatile oxidation products, with AsH_3 most rapidly oxidized and challenging to detect in environmental samples. Oxidation of the arsine gases to inorganic arsenic species completes the arsenic cycle, with arsenic returned to the soil by rain or dry deposition[101].

Arsenic methylation in soils increases with decreasing redox potential[102], and addition of organic matter. Increased arsenic volatilization was measured in soil after the addition of rice straw[103], and animal waste products[104]. Inoculation of fungi (*Penicillium* and *Ulocladium* spp.) increased arsenic volatilization up to 8 fold in heavily contaminated and spiked soils[105]. Microbially mediated arsenic volatilization remains very inefficient, which hinders attempts to use it in soil remediation. Gaseous arsines are volatilized from arsenic contaminated soils into the atmosphere at very low rates: a microcosm study found 0.5 – 70 μg of arsenic kg^{-1} soil year^{-1} was volatilized from a range of soils and a range of arsenic levels[27], and field measurements of arsenic volatilization are 1-2 orders of magnitude lower than those made in laboratory mesocosms [98].Genetic transformation of bacteria, using genes encoding for the protein product arsenite S-adenosyl methyltransferase (*ars*M) is an attempt to enhance arsenic methylation and volatilization. The *ars*M from *Rhodopseudomonas palustris* was expressed in *Sphingomonas desiccabilis* and *Bacillus idriensis* grown in an aqueous system, resulting in a 10-fold increase~~d~~ in arsenic volatilization compared to the wild type strains. In a soil-based system, 2.2 – 4.5% of arsenic was removed via microbially-mediated volatilization over an incubation period of 30 days[106] (See also section 5.3).

4.3. Changes in soil arsenic bioavailability due to ageing

Although arsenic in aerobic soils has a lower bioavailability and presents less of an immediate concern for crop uptake, aerobic soil can accumulate arsenic from human inputs, retain them for long periods of time, and release them when redox conditions change (See Section 3.3). Human inputs of arsenic, as discussed in Section 3, are diverse; biosolids, sewage sludge, coal fly ash, poultry litter, industrial waste, arsenical pesticides and from irrigation with naturally arsenic-enriched groundwater. For aerobic soils, ageing – where binding stability of arsenic to soil particles increases over time, is a particularly important part of arsenic cycling. Factors controlling ageing of arsenic include soil type, organic matter content and arsenic species. Both inorganic and organic arsenic species are subject to ageing, with studies indicating a slow oxidation process from arsenite to arsenate over time[107].

5. Rhizosphere processes

Processes occurring in the rhizosphere (the boundary layer of soil under the influence of plant roots) dramatically influence arsenic concentrations and bioavailability because they involve local alterations in redox potential, pH and organic matter content. Rhizosphere acidification occurs during iron uptake by all plant species ~~except grasses, whereas grasses secrete chelating agents (phytosiderophores) to acquire iron~~~~[96]~~during cation uptake and charge balance, when protons are released into the rhizosphere. Plants release anywhere from 10 to 250 mg of carbon per gram of root tissue into the rhizosphere; about 10-40% of their total photosynthetically fixed carbon[108], making the rhizosphere particularly rich in organic carbon compared to bulk soil, which in turn exerts an influence on arsenic solubility by stimulating microbially-mediated reductive dissolution of soil minerals. Large differences have been found in the arsenic concentration of rhizosphere soils compared with bulk soils in highly arsenic-contaminated areas, with higher concentrations of arsenic in rhizosphere soils compared to bulk soils[109].

In anaerobic soils, the iron plaque that develops on the submerged stem and roots of rice plants dominates rhizosphere dynamics of arsenic. In flooded environments such as paddy fields, plants oxygenate the rhizosphere through specialized tissues called aerenchyma, which are found in many aquatic plants and emergent macrophysics such as rice. This radial oxygen loss creates an oxidized layer around plant tissue that stimulates aerobic microbial activity and the oxidation of iron, which precipitates and forms a visible iron plaque on the root surface [110-114]. Formation of an iron (oxyhydr)oxide plaque on root surfaces can alter the uptake of arsenic by rice, acting as a sorbent for excess nutrients such as ferrous iron (reduced iron) as well as arsenic and aluminum[115]. Rates of oxygen loss influence iron plaque formation [115], and vary between rice cultivars[116, 117]. ~~However, s~~Studies conducted over the last forty years are inconsistent on whether iron plaque prevents or enhances arsenic uptake by plants[111], and the hypothesis that arsenic influences the quality and amount of the iron plaque[113]. Profound differences in mineral composition and quantity of laboratory-created iron plaques has been demonstrated experimentally[111], which may have contributed to these inconsistencies.

5.1. Microbial activity

Microbes directly and indirectly influence arsenic speciation in rhizosphere soil, and are widely considered to play a key role in arsenic biogeochemistry[118]. Under certain nutrient-limited conditions, microbes actively weather minerals to access nutrients for cellular growth, which releases arsenic[35], as well as creating abiotic conditions that induce changes in arsenic speciation via production of organic acids, polysaccharides and ligands. Soil microorganisms can strongly affect soil redox, regulating arsenic release into pore water[119]. A number of strains of bacteria have also been shown to contribute to the formation of arsenic minerals by using arsenic as a terminal electron acceptor, such as *Desulfosporosinus auripigmentum*[120], *Desulfovibrio* strain Ben-RB[121], *Shewanella oneidensis*[122] and *S. putrefaciences* CN32[123]. These microorganisms also differ in their capabilities for liberating arsenic from specific arsenate-bearing minerals[119].

Microbial transformation can mobilize arsenic by converting inorganic to organic forms, including MMA and DMA[124, 125]. Plants translocate organic arsenicals from roots to the (frequently edible) above-ground parts more efficiently than inorganic arsenic[126-128] (See Section 6), therefore microbial transformation to organic arsenicals can increase human dietary exposure.

Plants, green algae and microbes can all enzymatically transform arsenic species[124, 129], but methylated forms of arsenic detected in plants are a product of rhizosphere bacteria; plants cannot methylate arsenic[78, 124, 130, 131]. The genomes of more than 85 arsenic-metabolizing archaea and bacteria have been sequenced for genes involved in arsenic metabolism[132]. In bacteria, archea and fungi, arsenic methylation is catalyzed by homologs of arsM, (See Section 4)[124]. Resistance to arsenite and arsenate exists in nearly all microbes, which also confers the ability to transform arsenate into volatile arsine gases[133], a particularly effective way of removing arsenic.

Profiling the transcriptome, proteome and metabolome of arsenic contaminated soils offers way of understanding microbially-mediated rhizosphere arsenic processes[132]. This approach measures the presence and expression of specific genes, rather than attempting to isolate and study the microbes that carry them, 98% of which – it is estimated - do not grow in culture[134]. Microbially mediated arsenic metabolic processes that play a major role in arsenic cycling in agronomic systems include arsenite oxidation (via the *aio* genes), arsenate respiration (via the *arr* genes), arsenate reduction (via the *ars* genes) and arsenite methylation (via the *arsM* genes)[135]. Interested readers are referred to the recent excellent work of Andres and Bertin[132] for a comprehensive review of this subject. Microbially mediated redox processes strongly influence arsenic uptake in rice, involving *aioA*, *arsC* and *arrA* [124], with pH emerging as an important factor in the distribution of microbes in paddy soils. Testing a variety of soils has shown that bacteria possessing the *arsM* gene for methylating arsenic are widespread and phylogenetically diverse, and even in paddy soils with low concentrations of arsenic, genes for arsenic metabolism are abundant[124].

6. Arsenic and crop plants

Much of our understanding about the physiological mechanisms of arsenic uptake in plants comes from the study of a limited number of ~~plant~~ species. Called *model* *plant* species, they are extensively studied, well described, easy to grow, and the results can be compared between studies. The understanding is that the information gained from studying model plants is applicable to other plant species. From a genetic perspective, orthologous genes exist in different plant species having evolved from a common ancestral gene, and they usually retain the same function. Characterization of arsenic-related genes in a model plant strongly suggests that they exist and perform similar functions in other species. Caveats to this are their levels of expression, which makes some plants more adept at accumulating arsenic than others. In this section, much of the knowledge gained on arsenic uptake and metabolism of plants comes from the study of mouse-eared pennycress (also called thale cress or rockcress) (*Arabidopsis thaliana* Heynh.*)* and rice (*Oryza sativa* L.); model plants with fully sequenced genomes. These species represent dicotyledonous (e.g. flowers, vegetables, deciduous trees) and monocotyledonous plant species (e.g. grasses, palm trees) respectively, thereby representing much of the edible crop species. An exception to this is the study of the arsenic hyperaccumulating fern (Chinese Brake fern, *Pteris vittata*), a seedless plant that is able to accumulate up to 22,630 mg/kg (dry weight) arsenic in its fronds[136].

6.1. Phytotoxicity of arsenicals

Arsenic is toxic to plants[137]. Despite lower acute human toxicity of the organic arsenicals (median lethal dose is 700-1,600 mg/kg and 700-2,600 mg/kg for MMA and DMA respectively compared to 10-20 mg/kg for inorganic forms)[138] no one form of arsenic is consistently more toxic to plants[139]. Soybean yields are affected when tissue arsenic levels exceed 1 mg/kg, and 4 mg/kg limits cotton yields[140], whereas in barley tissue concentrations of 20 mg/kg inhibited growth[141]. Higher yield-limiting arsenic levels have been recorded in rice: 20-100 mg/kg in above ground biomass, and 1000 mg/kg in root tissue[142]. By contrast, potatoes (*Solanum tuberosum* L.) suffered no growth inhibition in soils containing 290 mg/kg arsenic[71]. In some plants species, organic forms are more toxic than inorganic, for example in rice (order of toxicity: MMA > arsenite > arsenate = DMA)[143], and in smooth cordgrass (*Spartina alterniflora* Loisel) (DMA = MMA > arsenite > arsenate)[144].

Plants vary in their tolerance to arsenic, and the stress response differs for each arsenic species[145-147]. The chemical similarities between arsenate and phosphate means that arsenic can replace phosphate in biomolecules like ATP (adenosine triphosphate, a molecule used for intercellular energy transfer), with negative impacts on growth and metabolism[148]. In rice in particular, DMA and MMA induce straighthead disease (arsenic-associated straighthead disease), significantly lowering yield of certain rice varieties[66]. Straighthead is a physiological disorder of rice characterized by sterile florets, which remain upright at maturity instead of bending over under the weight of the filled grain. The exact cause of straighthead is unknown, but consistent flooding, low soil pH, high iron availability and high organic matter content have all been implicated in naturally-occurring straighthead disease[66]. Arsenic's suspected role in straighthead comes from observations of more frequent outbreaks in rice grown in soil where arsenical herbicides such as monosodium methanearsonate (MSMA) – used in cotton production in the USA – have been historically applied.

6.2. Arsenic uptake mechanisms

In magnitude, plants take up arsenicals from the soil in the order arsenite > arsenate > DMA > MMA[149, 150)], with the various arsenic species entering via different root membrane transport proteins in the root plasma membrane that allow ions and molecules to cross with varying levels of selectivity, or target specificity. Similarities in chemical structure between arsenate and phosphate, and between arsenite and silicic acid, govern their entry into root cells. Arsenate enters root cells through phosphate transporters (the Phosphate Transporter 1 family of proteins; PHT1) in both the model plant *Arabidopsis thaliana*[151, 152] and in rice[153-155] (Figure 1). In rice, Low Silicon 1 (OsLsi1) and OsLsi2 are silicic acid transporters and arsenite, MMA^{V}, and DMA^{V} are among their unintended targets[156, 157]. ~~The transporter that allows arsenite initial entry to root cells remains unidentified, but is hypothesized to be a member of the Plasma Membrane Intrinsic Protein family. Inside the root,~~These Nodulin 26-like Intrinsic Proteins (NIPs)[158], which are members of the aquaporin water channel superfamily of proteins[159] embedded in the exodermal cell membranes of rice roots, move arsenic from the soil into the vascular system for distribution to the stem and leaves. ~~In rice, Low Silicon 1 (OsLsi1) and OsLsi2 are silicic acid transporters and arsenite, MMA^{V}, and DMA^{V} are among their unintended targets[148, 149].~~ OsLsi2 works in tandem with OsLsi1 to transport arsenite inward toward the xylem[160, 161] (vascular tissue that conducts water and dissolved nutrients up from the roots). The arsenic uptake

Field Code Changed

Field Code Changed

specificity of OsLsi1 is arsenite >> MMA > DMA[158]. These bidirectional NIP transport proteins also efflux arsenite back in to the soil, but since OsLsi1 effluxes only 15-20% of the arsenite in roots cells[162], there may be other unidentified arsenite efflux transporters contributing to this process.

6.3. Arsenic transport and metabolism in plants

Transport of arsenite into the xylem for delivery to the shoot is less well characterized than its uptake from the soil. Arsenic is transported to the grain mainly via the phloem[126-128] (vascular tissue that conducts sugars and metabolic products from the leaves), by transporters in the nodes[163], but their characterization is still in the early stages. Transporters for myo-inositol (Inositol Transporter 2 and 4); an important sugar for developing rice grains, also transport arsenite into the phloem companion cells[164, 165]. In Arabidopsis, ~~At~~INT2 or AtINT4 load about 45-64% arsenite into the grain[166]. The identity of transporters that move arsenite out of the phloem and into the grain are also unknown, but manipulating the target specificity of the INT genes might show promise in molecular genetic or plant breeding mitigation efforts as a way to prevent arsenite from reaching the grain.

~~Once inside the cell~~Despite having a lower affinity for transporters into the plant than the inorganic forms, ~~the rate of arsenic transport up towards shoot tissue is reversed, with~~ organic ~~forms~~ arsenic species are more efficiently transported towards the shoot than inorganic forms[149, 150] because they are not complexed by phytochelatins (PCs); sulphydryl-rich glutathione (GSH) polymers[167, 168]. Likewise, in broad beans (*Vicia faba*

L.) grown in a soil containing 90% inorganic arsenic, DMA and MMA were the dominant arsenic forms in the bean (68%)[169]. In root vegetables, carrot (*Daucus carota* L.) and beet (*Beta vulgaris* L.) grown on arsenic-contaminated soils, arsenic forms were predominantly inorganic, ~~and~~ but for beets in particular were not readily identified using the typical standards (arsenate, arsenite, MMA and DMA)[170].

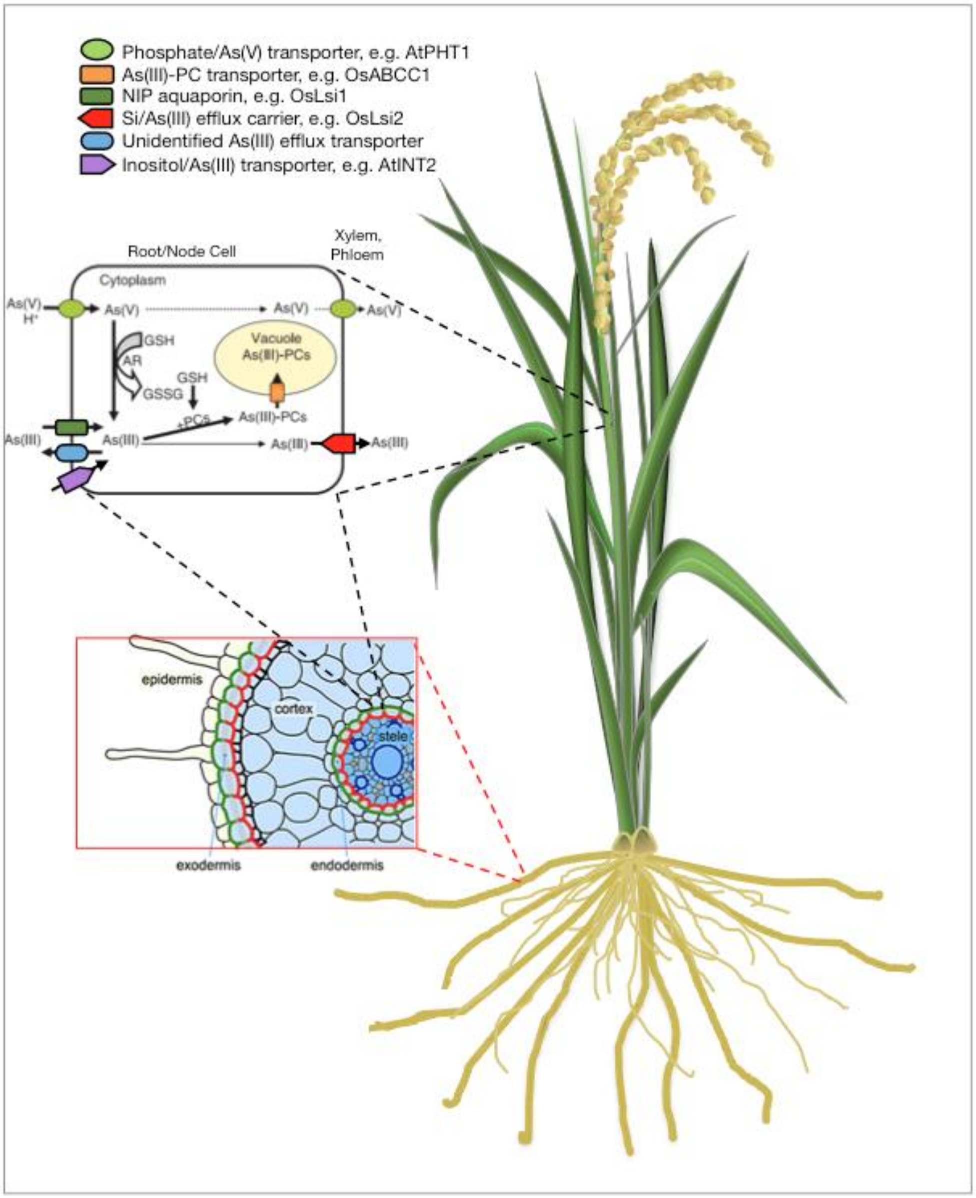


Figure 1. Generalized diagram of arsenic uptake, transport and metabolism in plants. GSH, glutathione; AR, arsenate reductase; GSSG, oxidized glutathione; PC, phytochelatin. Modified from Zhao et al.[171] and Ma et al.[172].

The arsenic species composition of rice grain is influenced by the arsenic transport rate of the particular cultivar[173, 174]. Rice cultivars currently grown in the USA have an arsenic speciation split approximately equally between inorganic arsenic and DMA, while cultivars grown in Bangladesh contain mostly inorganic arsenic[174]. While lower inorganic arsenic in rice grain seems favorable for avoiding human health effects, the assumed safety of DMA is contentious[175], being based on acute toxicity data, and not on genotoxicity or carcinogenicity, which are equally relevant in long term safety considerations.

Arsenic detoxification inside cells uses a multi-step process beginning with reduction of arsenate to arsenite using an arsenate reductase enzyme[176, 177]. In Arabidopsis, the protein High Arsenic Content1 (~~At~~HAC1; also called Arsenate Reductase QTL1; ARQ1) reduces arsenate[177]. Even though arsenite is more toxic than arsenate[158, 178, 179], it is hypothesized that ancestral organisms to plants were exposed almost exclusively to arsenite before atmospheric oxygen enabled arsenate formation[180], and this mechanism persisted through natural selection. Arsenite is then complexed by PCs, and transported in to the vacuole[167] via ATP Binding Cassette~~-type~~ tranporters[181, 182]. This process depletes glutathione availability, rendering the plant more susceptible to other oxidative stresses, which ~~negatively impact~~inhibits photosynthesis, pigment production, and the integrity of cell membranes[183-186].

7. Limiting arsenic uptake by crops

7.1. Water management

Although the traditional method for cultivating rice involves flooding leveled, tilled fields before or shortly after planting germinated seedlings, ~~this~~ flooded soil is not a biological requirement of rice plants. Flooding is used for weed and vermin control, for mobilization of key nutrients such as iron, phosphate and zinc, and importantly, flooding discourages the buildup of root nematodes over multiple years of rice growth. As mentioned earlier, flooded conditions mobilize soil-bound arsenic through reductive dissolution of Fe (oxyhydr)oxides, and the reduction of arsenate to the more mobile arsenite[187]. Water management strategies that involve periods of oxic soil conditions can decrease arsenic uptake in rice by limiting dissolution of arsenic. Rice grown in non-flooded or aerobic conditions has a lower yield than intermittently or constantly flooded rice[188-190]. Intermittent flooding (flooding maintained until full tillering, followed by intermittent irrigation) is a promising management technique to reduce arsenic levels, and can potentially produce higher grain yields than either non-flooded or constantly flooded conditions[191]. However, oxic conditions increase cadmium concentrations in the grain when grown in acidic soils[191-193], and cadmium is also a highly toxic metal. The observed increases in cadmium were also a cultivar-specific trait, but the increase in cadmium uptake between rice grown under aerobic conditions were approximately an order of magnitude greater than their flooded counterparts. Pot experiments suggest that water management strategies implemented during the heading period of rice growth (when the rice panicle has emerged from the stem and is fully visible, just before flowering) can regulate both arsenic and cadmium concentration in the grain[190, 192].

7.2. Amendment and fertilization practices

Soil amendment involves incorporating substances into the plow layer that either add missing nutrients, reduce the bioavailability of existing potentially toxic substances (to prevent crop uptake), or both. Soil amendments that have shown potential in reducing arsenic uptake by plants include iron-, and silica-based additives. The use of iron-based amendments increases in the concentration of free iron oxide in the soil, retarding the release of arsenite from the solid phase into soil solution, (mentioned in Section 4.1 and discussed in Section 5), whereas silica fertilization, inhibits arsenic uptake by competitive inhibition at the plant root surface while adding an essential nutrient.

Zero valent iron powder (90% iron) and iron oxide (56% iron) incorporation prevented uptake of arsenic in to the grain of rice grown on soil containing 39.5 mg/kg total arsenic by approximately 45%, and corresponded with a reduction in bioavailable arsenic in the soil[194]. Amendment with iron oxides (at a rate of 2%) was also more effective at reducing grain arsenic than phosphate amendment[195]. Amendments have also been used in combination with water management strategies to try and reduce both arsenic and cadmium concentrations in rice simultaneously[193], without success. Reduction of arsenic in the grain was achieved with iron oxide addition and constant flooding, whereas cadmium reduction was achieved with converter furnace slag addition and rain water management (no irrigation after midseason drainage until harvest).

A combination of ethylenediaminetetraceetic acid ferric sodium salt (iron EDTA) and calcium peroxide was effective for reducing arsenic uptake by vegetable crops (lettuce, Chinese cabbage and radish) from soils containing 14 mg/kg total arsenic[196], again by increasing amorphous aluminum and iron oxides. It is likely that this this level of arsenic contamination would be deemed too high for commercial vegetable production, so these amendments may only be feasible for use in private vegetable gardens. Questions remain about whether iron oxide amendment application only temporarily reduces arsenic bioavailability[197]. In addition, the suitability for arsenic immobilization is highest at the lower soil pH range, and is strongly affected by soil phosphorus concentration, which strongly competes with arsenic.

Rice plants take up high concentrations of silica, constituting up to 10% of dry matter in the straw and husk of the plant[198]. As mentioned earlier (Section 6) the silicon membrane transporter (Lsi1) is the main route of arsenite entry in to rice root cells, and provision of silicon causes competitive inhibition of arsenite uptake. Increasing silicon availability in the soil also reduces the expression of the Lsi1 transporter in the plant, which further decreases the potential for arsenic uptake. ~~.~~ Fertilization of rice paddy soils with silicon is a potential mitigation strategy for preventing or reducing arsenic uptake by rice through competitive inhibition of arsenite uptake[199]. The use of synthetic silicon fertilizers, such as calcium silicate or silica gel is prohibitively expensive for smallholder farmers in developing countries, however reusing the silicon-rich parts of the rice plant that remain after harvesting and grain processing may provide a sustainable solution that also addresses the ongoing issue of silicon depletion of the soil[198]. Soil incorporation of fresh rice husks, or the ash that remains after burning the husk and straw for energy (which is a common practice ~~in small holding farms~~for smallholder farmers), can provide silicon without increasing ~~arsenic or~~ methane production and decreases either total or inorganic arsenic in rice grain[200].

Despite the potential of soil amendment with iron oxides, or silica to reduce arsenic bioavailability or prevent plant uptake of arsenic, the high cost of these amendments inevitably prevents their use, especially by smallholder farmers. Large rice producers in the US or Europe have not so far adopted widespread use of these soil amendments to reduce rice grain arsenic concentrations. It is reasonable to assume that use of such costly amendments would also drive up the cost of rice. It may also be that iron amendments are essentially untested in a diverse range of large-scale agricultural settings and their performance will vary from soil to soil. In non-rice agricultural systems arsenic is tightly bound to the solid phase; significant crop uptake from oxidized soil is likely to be a result of extreme contamination, in which case effective mitigation is restricted to redirecting land use away from edible crops. In systems subject to periodic flooding, improving drainage remains the best mitigation strategy.

7.3. Mitigation using plant breeding approaches

The development of crops that accumulate high levels of arsenic and yet remain healthy, while preventing arsenic from reaching the edible grain is thought to hold great potential as a strategy for reducing human exposure to dietary arsenic. The use of molecular genetics techniques such as alterations in gene expression characteristics, gene editing to alter target specificity, or alternately, using traditional plant breeding techniques are both tangible approaches. Both use knowledge of the arsenic uptake and tolerance characteristics of plants to develop varieties with desired characteristics. These characteristics include lower arsenic uptake[201], higher arsenite efflux[202] and increased vacuolar arsenic sequestration[203]. For instance, many rice cultivars have now been screened to identify those that accumulate lower levels of arsenic in their grain and efforts are underway to identify the genes underlying this trait[201, 204]. Overexpressing Arabidopsis ABC-type transporters that sequester arsenite-PC complexes in the cell vacuole results in plants able to grow in otherwise toxic concentrations of arsenic[182]. Conversely, knocking out the function of the related rice ABC transporter OsABCC1 results in higher levels of grain arsenic. The OsABCC1 transporter limits arsenic transport to grains by sequestering arsenic in the vacuoles of the phloem companion cells directly connected to the grain. By combining what we have learned from the overexpression studies in Arabidopsis and the loss-of-function study in rice, overexpression of OsABCC1 can be used as a strategy to breed arsenic tolerance and low-arsenic accumulating rice cultivars. Another promising strategy is based on expressing the arsenate efflux transporter from yeast (*Saccharomyces cerevisiae*) in rice, which can reduce arsenic accumulation in brown rice by 20%. A less successful idea to methylate sodium arsenite to DMA by expressing an algal arsM gene in Arabidopsis resulted in lethal phytotoxicity[205], suggesting that arsenic methylation in plants can only be an effective detoxification strategy if volatile arsines ~~were~~ are the end point of the methylation.

8. Conclusions

The discovery of arsenic in staple foods, beverages and other products has increased awareness and stimulated research on the sources and the processes involved. The information brought together here illustrates the ~~wide diversity of natural and human processes that can contribute arsenic to the soil and the~~ numerous geochemical and biological processes that ~~can move~~influence the movement of arsenic into the food supply.

It is clear there must be strategies for ~~protecting~~ preventing ~~human health to~~ arsenic exposure, that operate in both the short term – to protect consumers from existing contamination – and in the long term,~~;~~ to prevent further contamination. This requires government regulation on the permissible levels of arsenic food, with lower levels for infant foods (see Nachman et al, this issue), which must work in tandem with long term goals to ~~remediate~~ address arsenic in agricultural soils, actively prevent further inputs and identify contaminated areas. Our recommendations are that the information in this review is used to inform a reconsideration and a unification of regulations on the action levels of agricultural soil arsenic, which in the USA for example, exist only at the state level, vary widely from state to state, and have no formal channels of enforcement. We recommend that educating the community and garnering their support and involvement for lowering exposure to arsenic through food is an approach already shown to hold enormous potential.

Direct involvement of the commercial rice growing community in research and development of arsenic mitigation strategies and amendments is needed. Community-based participatory research should extend to the agricultural community, leading to partnerships that will make longer-term field-scale testing accessible.

Much effort is given to short-term, greenhouse-scale testing of amendment formulations that will ultimately be too expensive, impractical, or ineffective in the long term. Feasibility should be the first consideration in arsenic mitigation research. Community outreach efforts targeted to commercial growers or the home gardener specifically ~~could~~ must raise awareness of the significance and potential impacts of the former land uses ~~of the land~~, encouraging testing for the presence of arsenic in the soil and educating growers on the kinds of crops shown to accumulate arsenic in their edible parts. Information gathering on former arsenic input into the soil from pesticides and from proximity to various waste sites is of paramount importance ~~to this effort~~, and will allow monitoring to be targeted to where it is needed most. This information needs to be accessible to everyone, everywhere; currently there is no readily available source of soil quality information at a sufficient resolution to inform commercial producers or homeowners. Going forward, management and remediation of arsenic contaminated soils is essential both for human health and food security, and innovative technologies are urgently needed that will expedite this process. Innovative solutions such as the use of rice husks to add silicon to the soil to offset arsenic uptake, and the use of cultivars with low-arsenic accumulating characteristics point the way forward for sustainable solutions.

Acknowledgements

This paper, a product of the Collaborative on Food with Arsenic and associated Risk and Regulation (C-FARR), is supported by the Dartmouth College Toxic Metals Superfund Research Program through funds from the National Institute of

Environmental Health Sciences of the National Institutes of Health under Award Number 1R13ES026493-01 to C. Chen and Award Number P42ES007373 to B. Stanton, and the Children's Environmental Health and Disease Prevention Research Center at Dartmouth through funds from the National Institute of Environmental Health Sciences of the National Institutes of Health under Award Number P01ES022832 to M. Karagas. Although EPA contributed to this article, the research presented was not performed by or funded by EPA and was not subject to EPA's quality system requirements. Consequently, the views, interpretations, and conclusions expressed in this article are solely those of the authors and do not necessarily reflect or represent EPA's views or policies. TP was supported by funds from the National Institute of General Medical Sciences Center for Biomedical Research Excellence (P20GM104416). The authors disclose that there are no actual or potential conflicts of interest.

Bibliography

1. Norton, G.J., et al., *Variation in grain arsenic assessed in a diverse panel of rice (Oryza sativa) grown in multiple sites.* New Phytologist, 2012. **193**(3): p. 650-664.
2. Carlin, D.J., et al., *Arsenic and Environmental Health: State of the Science and Future Research Opportunities.* Environ Health Perspect, 2015.
3. Bodwell, J.E., L.A. Kingsley, and J.W. Hamilton, *Arsenic at very low concentrations alters glucocorticoid receptor (GR)-mediated gene activation but not GR mediated gene repression; complex dose response effects are closely correlated with levels of activated GR and require a functional GR DNA binding domain.* Chemical Research in Toxicology, 2004. **17**: p. 1064-1076.
4. Davis, M.A., et al., *Rice consumption and urinary arsenic concentrations in US children.* Environmental Health Perpectives, 2012. **120**: p. 1418–1424.
5. WHO. *Arsenic*. 2016 [cited 2016 November 22, 2016]; International Programme on Chemical Safety. Health Impacts of Chemicals:]. Available from: http://www.who.int/ipcs/assessment/public_health/arsenic/en/.
6. Union, E. *Commission Regulation (EU) 2015/1006 of 25 June 2015 amending Regulation No. 1881/2006 as regards maximum levels of inorganic arsenic in foodstuffs*. 2015 [cited 2016 November 22, 2016]; Free access to EU law (treaties, directives, regulations, decisions, consolidated legislation), preparatory acts, case-law, international agreements, EFTA focuments, summaries of EU legislation and other documents.]. Available from: http://eur-lex.europa.eu/eli/reg/2015/1006/oj.
7. Moon, K., E. Guallar, and A. Navas-Acien, *Arsenic exposure and cardiovascular disease: an updated systematic review.* Curr Atheroscler Rep, 2012. **14**(6): p. 542-55.
8. Caldwell, K.E., et al., *Prenatal arsenic exposure alters the programming of the glucocorticoid signaling system during embryonic development.* Neurotoxicol Teratol, 2014. **47C**: p. 66-79.
9. Yamamura, S., et al., *Drinking water guidelines and standards. In United Nations Synthesis Report on Arsenic in Drinking water.* . 2001, United Nations: United Nation, Geneva. p. 18.

10. FDA, U., *FDA proposes limit for inorganic arsenic in infant rice cereal*, L. Sucher, Editor. 2016, US Food and Drug Administration.

11. Japan, M.o.E.G.o. *Environmental Quality Standards for Soil Pollution*. Water/Soil/Ground Environment 2016 [cited 2016 12 December].

12. Henke, K., *Appendix E: Regulation of Arsenic: A Brief Survey and Bibliography*, in *Arsenic*, K. Henke, Editor. 2009, John Wiley & Sons, Ltd. p. 545-557.

13. Adomako, E.E., et al., *Enhanced transfer of arsenic to grain for Bangladesh grown rice compared to US and EU.* Environment International, 2009. **35**(3): p. 476-479.

14. Lu, Y., et al., *Baseline Soil Variation Is a Major Factor in Arsenic Accumulation in Bengal Delta Paddy Rice.* Environmental Science & Technology, 2009. **43**(6): p. 1724-1729.

15. Williams, P.N., et al., *Greatly enhanced arsenic shoot assimilation in rice leads to elevated grain levels compared to wheat and barley.* Environmental Science & Technology, 2007. **41**(19): p. 6854-6859.

16. Norton, G., et al., *Arsenic Speciation and Localization in Horticultural Produce Grown in a Historically Impacted Mining Region.* Environmental Science & Technology, 2013. **47**(12): p. 6164-6172.

17. Koljonen, T., et al., *12th International Geochemical Exploration Symposium and the 4th Symposium on Methods of Geochemical Prospecting Geochemical Atlas of Finland: preliminary aspects.* Journal of Geochemical Exploration, 1989. **32**(1): p. 231-242.

18. Geiszinger, A., W. Goessler, and W. Kosmus, *Organoarsenic compounds in plants and soil on top of an ore vein.* Applied Organometallic Chemistry, 2002. **16**(5): p. 245-249.

19. IMA, *The new International Mineralogical Association list of minerals. A work in progress.* . 2014, International Mineralogical Society: http://www.ima-mineralogy.org/Minlist.htm.

20. Lado, L.R., T. Hengl, and H.I. Reuter, *Heavy metals in European soils: A geostatistical analysis of the FOREGS Geochemical database.* Geoderma, 2008. **148**(2): p. 189-199.

21. Shacklette, H.T. and J.G. Boerngen, *Element Concentrations in soils and other surficial materials of the Conterminous United States*. 1984, US Geological Survey: US Government Printing Office, Washington, DC. p. 105.

22. Smith, D.B., et al., *Geochemical and mineralogical maps for soils of the conterminous United States: U.S.*, U.G. Survey, Editor. 2014. p. 386.

23. USGS. *Mineral Resources On-Line Spatial Data: National Geochemical Survey database*. [Website] 2016 September 3 2014 [cited 2016 July 18]; National scale geochemical analysis of stream sediments and soils in the US, from existing data, reanalysis of existing samples and new sampling.]. Available from: http://mrdata.usgs.gov/geochemistry/ngs.html.

24. Young, M. and A. Donald, *A guide to the Tellus data.* 2013, Belfast, UK: Geological Survey of Northern Ireland. 233.

25. Smedley, P.L. and D.G. Kinniburgh, *A review of the source, behaviour and distribution of arsenic in natural waters.* Applied Geochemistry, 2002. **17**(5): p. 517-568.

26. Saunders, J.A., et al., *Natural arsenic contamination of Holocene alluvial aquifers by linked tectonic, weathering, and microbial processes.* Geochemistry Geophysics Geosystems, 2005. **6**.

27. Mestrot, A., et al., *Field Fluxes and Speciation of Arsines Emanating from Soils.* Environmental Science & Technology, 2011. **45**(5): p. 1798-1804.

28. Lawgali, Y.F. and A.A. Meharg, *Levels of Arsenic and Other Trace Elements in Southern Libyan Agricultural Irrigated Soil and Non-irrigated Soil Projects.* Water Quality Exposure and Health, 2011. **3**(2): p. 79-90.

29. Melegy, A., et al., *Weathering fluxes of arsenic from a small catchment in Slovak Republic.* Environmental Earth Sciences, 2011. **64**(2): p. 549-555.

30. Drahota, P., et al., *Weathering and erosion fluxes of arsenic in watershed mass budgets.* Science of the Total Environment, 2006. **372**(1): p. 306-316.

31. Huang, J.H. and E. Matzner, *Biogeochemistry of organic and inorganic arsenic species in a forested catchment in Germany.* Environmental Science & Technology, 2007. **41**(5): p. 1564-1569.

32. CEH. *United Kingdom Pollutant Deposition Maps*. 2008 2016]; Database]. Available from: http://www.pollutantdeposition.ceh.ac.uk/content/heavy-metals.

33. UKSO. *NSI topsoil Arsenic map*. 2016 [cited 2016 August 11]; Topsoil arsenic map of England and Wales]. Available from: http://www.ukso.org/nsi/Arsenic.html.

34. Mikutta, C. and J.J. Rothwell, *Peat Bogs as Hotspots for Organoarsenical Formation and Persistence.* Environmental Science & Technology, 2016. **50**(8): p. 4314-4323.

35. Mailloux, B.J., et al., *Microbial Mineral Weathering for Nutrient Acquisition Releases Arsenic.* Applied and Environmental Microbiology, 2009. **75**(8): p. 2558-2565.

36. Williams, P.N., et al., *Organic Matter-Solid Phase Interactions Are Critical for Predicting Arsenic Release and Plant Uptake in Bangladesh Paddy Soils.* Environmental Science & Technology, 2011. **45**(14): p. 6080-6087.

37. Murphy, E.A. and M. Aucott, *An assessment of the amounts of arsenical pesticides used historically in a geographical area.* The Science of The Total Environment, 1998. **218**(2-3): p. 89-101.

38. Lasky, T., et al., *Mean total arsenic concentrations in chicken 1989-2000 and estimated exposures for consumers of chicken.* Environmental Health Perspectives, 2004. **112**(1): p. 18-21.

39. Nachman, K.E., et al., *Roxarsone, Inorganic Arsenic, and Other Arsenic Species in Chicken: A US-Based Market Basket Sample.* Environmental Health Perspectives, 2013. **121**(7): p. 818-824.

40. Fendorf, S. and B.D. Kocar, *Chapter 3 Biogeochemical Processes Controlling the Fate and Transport of Arsenic: Implications for South and Southeast Asia*, in *Advances in Agronomy*. 2009, Academic Press. p. 137-164.

41. Bowell, R.J., et al., *The Environmental Geochemistry of Arsenic - An Overview -.* Arsenic: Environmental Geochemistry, Mineralogy, and Microbiology, 2014. **79**: p. 1-16.

42. Craw, D. and R.J. Bowell, *The Characterization of Arsenic in Mine Waste*, in *Arsenic: Environmental Geochemistry, Mineralogy, and Microbiology*, R.J. Bowell, et al., Editors. 2014. p. 473-505.

43. Bowell, R.J., et al., Reviews in mineralogy and geochemistry, 2014. **79**: p. 635.

44. Bowell, R.J., et al., *The Environmental Geochemistry of Arsenic - An Overview*, in *Arsenic: Environmental Geochemistry, Mineralogy, and Microbiology*, R.J. Bowell, et al., Editors. 2014. p. 1-16.

45. Ramirez-Andreotta, M.D., et al., *Home gardening near a mining site in an arsenic-endemic region of Arizona: Assessing arsenic exposure dose and risk*

via ingestion of home garden vegetables, soils, and water. Science of the Total Environment, 2013. **454**: p. 373-382.

46. Ramirez-Andreotta, M.D., et al., *A greenhouse and field-based study to determine the accumulation of arsenic in common homegrown vegetables grown in mining-affected soils.* Science of the Total Environment, 2013. **443**: p. 299-306.
47. Bertoldi, D., et al., *Arsenic present in the soil-vine-wine chain in vineyards situated in an old mining area in Trentino, Italy.* Environmental Toxicology and Chemistry, 2013. **32**(4): p. 773-779.
48. Ma, L., et al., *Arsenic speciation in locally grown rice grains from Hunan Province, China: Spatial distribution and potential health risk.* Science of The Total Environment, 2016. **557–558**: p. 438-444.
49. Williams, P.N., et al., *Occurrence and Partitioning of Cadmium, Arsenic and Lead in Mine Impacted Paddy Rice: Hunan, China.* Environmental Science & Technology, 2009. **43**(3): p. 637-642.
50. Zhu, Y.G., et al., *High percentage inorganic arsenic content of mining impacted and nonimpacted Chinese rice.* Environmental Science & Technology, 2008. **42**(13): p. 5008-5013.
51. Kolker, A., et al., *Arsenic in coal*, in *Fact Sheet*. 2006. p. 4.
52. EPA. *2016*. Coal Ash Basics [Web Page] EPA June 7, 2016; Available from: https://www.epa.gov/coalash/coal-ash-basics.
53. Feuerborn, H.-J. *Coal Combustion Products in Europe - an update on Production and Ultilisation Standardisation and Regulation.* in *World of Coal Ash (WOCA) Conference*. 2011. Denver, CO, USA.
54. Yager, J.W., T. Greene, and R.A. Schoof, *Arsenic relative bioavailability from diet and airborne exposures: Implications for risk assessment.* Science of the Total Environment, 2015. **536**: p. 368-381.
55. Schmidt, C., *COAL ASH SPILL STILL A PROBLEM.* Chemical & Engineering News Archive, 2010. **88**(49): p. 13.
56. Schumann, A.W. and M.E. Sumner, *Plant nutrient availability from mixtures of fly ashes and biosolids.* Journal of Environmental Quality, 1999. **28**(5): p. 1651-1657.

57. Jackson, B.P., et al., *Trace element solubility from land application of fly ash/organic waste mixtures.* Journal of Environmental Quality, 1999. **28**(2): p. 639-647.

58. Schumann, A.W. and M.E. Sumner, *Formulation of environmentally soundwaste mixtures for land application.* Water Air and Soil Pollution, 2004. **152**(1-4): p. 195-217.

59. Yokel, J. and D.A. Delistraty, *Arsenic, lead, and other trace elements in soils contaminated with pesticide residues at the Hanford site (USA).* Environmental Toxicology, 2003. **18**(2): p. 104-114.

60. Embrick, L.L., et al., *Characterization of lead and arsenic contamination at Barber Orchard, Haywood County, NC.* Microchemical Journal, 2005. **81**(1): p. 117-121.

61. Renshaw, C.E., et al., *Impact of land disturbance on the fate of arsenical pesticides.* Journal of Environmental Quality, 2006. **35**(1): p. 61-67.

62. Peryea, F.J. and T.L. Creger, *VERTICAL-DISTRIBUTION OF LEAD AND ARSENIC IN SOILS CONTAMINATED WITH LEAD ARSENATE PESTICIDE-RESIDUES.* Water Air and Soil Pollution, 1994. **78**(3-4): p. 297-306.

63. Robinson, G.R., et al., *Assessment of contamination from arsenical pesticide use on orchards in the Great Valley region, Virginia and West Virginia, USA.* Journal of Environmental Quality, 2007. **36**(3): p. 654-663.

64. Peryea, F.J. and R. Kammereck, *Phosphate-enhanced movement of arsenic out of lead arsenate-contaminated topsoil and through uncontaminated subsoil.* Water Air and Soil Pollution, 1997. **93**(1-4): p. 243-254.

65. Bednar, A.J., et al., *Presence of organoarsenicals used in cotton production in agricultural water and soil of the southern United States.* Journal of Agricultural and Food Chemistry, 2002. **50**(25): p. 7340-7344.

66. Rahman, M.A., et al., *Straighthead disease of rice (Oryza sativa L.) induced by arsenic toxicity.* Environmental and Experimental Botany, 2008. **62**(1): p. 54-59.

67. Hudak, P.F., *Distribution and sources of arsenic in the southern High Plains Aquifer, Texas, USA.* Journal of Environmental Science and Health Part a-Toxic/Hazardous Substances & Environmental Engineering, 2000. **35**(6): p. 899-913.

68. Reedy, R.C., et al., *Unsaturated zone arsenic distribution and implications for groundwater contamination.* Environmental Science & Technology, 2007. **41**(20): p. 6914-6919.
69. Ayotte, J.D., et al., *Modeling the probability of arsenic in groundwater in New England as a tool for exposure assessment.* Environmental Science & Technology, 2006. **40**(11): p. 3578-3585.
70. Feng, M., et al., *Arsenic transport and transformation associated with MSMA application on a golf course green.* Journal of Agricultural and Food Chemistry, 2005. **53**(9): p. 3556-3562.
71. Codling, E.E., R.L. Chaney, and C.E. Green, *Accumulation of Lead and Arsenic by Potato Grown on Lead-Arsenate-Contaminated Orchard Soils.* Communications in Soil Science and Plant Analysis, 2016. **47**(6): p. 799-807.
72. Codling, E.E., R.L. Chaney, and C.E. Green, *Accumulation of lead and arsenic by carrots grown on lead-arsenate contaminated orchard soils.* Journal of Plant Nutrition, 2015. **38**(4): p. 509-525.
73. McBride, M.B., et al., *Arsenic and Lead Uptake by Vegetable Crops Grown on an Old Orchard Site Amended with Compost.* Water Air and Soil Pollution, 2015. **226**(8).
74. Lim, M.P. and M.B. McBride, *Arsenic and lead uptake by Brassicas grown on an old orchard site.* Journal of Hazardous Materials, 2015. **299**: p. 656-663.
75. Zavala, Y.J. and J.M. Duxbury, *Arsenic in rice: I. Estimating normal levels of total arsenic in rice grain.* Environmental Science & Technology, 2008. **42**(10): p. 3856-60.
76. Zavala, Y.J., et al., *Arsenic in rice: II. Arsenic speciation in USA grain and implications for human health.* Environmental Science & Technology, 2008. **42**(10): p. 3861-6.
77. Williams, P.N., et al., *Variation in arsenic speciation and concentration in paddy rice related to dietary exposure*. Environmental Science and Technology, 2005. **39**(15): p. 5531-5540.
78. Lomax, C., et al., *Methylated arsenic species in plants originate from soil microorganisms.* New Phytol, 2012. **193**(3): p. 665-72.
79. Pouschat, P. and G.J. Zagury, *In vitro gastrointestinal bioavailability of arsenic in soils collected near CCA-treated utility poles.* Environmental Science & Technology, 2006. **40**(13): p. 4317-4323.

80. Townsend, T.G., et al., *Chromium, copper and arsenic concentrations in soil underneath CCA-treated wood structures. .* Soil & Sediment Contamination, 2003. **12**: p. 779-798.

81. Juhasz, A.L., J. Weber, and E. Smith, *Predicting Arsenic Relative Bioavailability in Contaminated Soils Using Meta Analysis and Relative Bioavailability-Bioaccessibility Regression Models.* Environmental Science & Technology, 2011. **45**(24): p. 10676-10683.

82. Carbonell-Barrachina, A.A., et al., *Arsenic chemistry in municipal sewage sludge as affected by redox potential and pH.* Water Research, 2000. **34**(1): p. 216-224.

83. USFDA. *Arsenic-based Animal Drugs and Poultry*. Available from: http://www.fda.gov/AnimalVeterinary/SafetyHealth/ProductSafetyInformation/ucm257540.htm.

84. Taylor, D.A., *Funky chicken - Consumers exposed to arsenic in poultry.* Environmental Health Perspectives, 2004. **112**(1): p. A50-A50.

85. Bednar, A.J., et al., *Photodegradation of roxarsone in poultry litter leachates.* The Science of The Total Environment, 2003. **302**(1-3): p. 237-245.

86. Stolz, J.F., et al., *Biotransformation of 3-nitro-4-hydroxybenzene arsonic acid (roxarsone) and release of inorganic arsenic by Clostridium species.* Environmental Science & Technology, 2007. **41**(3): p. 818-823.

87. Garbarino, J.R., et al., *Environmental fate of roxarsone in poultry litter. I. Degradation of roxarsone during composting*. Environmental Science & Technology, 2003. **37**(8): p. 1509-1514.

88. Jackson, B.P., et al., *Trace element speciation in poultry litter.* J Environ Qual, 2003. **32**(2): p. 535-40.

89. Jackson, B.P., J.C. Seaman, and P.M. Bertsch, *Fate of arsenic compounds in poultry litter upon land application.* Chemosphere, 2006. **65**(11): p. 2028-2034.

90. Rutherford, D.W., et al., *Environmental fate of roxarsone in poultry litter. part II. Mobility of arsenic in soils amended with poultry litter.* Environmental Science & Technology, 2003. **37**(8): p. 1515-1520.

91. Han, F.X., et al., *Arsenic solubility and distribution in poultry waste and long-term amended soil.* Science of the Total Environment, 2004. **320**(1): p. 51-61.

92. Fisher, D.J., L.T. Yonkos, and K.W. Staver, *Environmental Concerns of Roxarsone in Broiler Poultry Feed and Litter in Maryland, USA.* Environmental Science & Technology, 2015. **49**(4): p. 1999-2012.

93. Shah, S.B., et al., *Leaching of Nutrients and Trace Elements from Stockpiled Turkey Litter into Soil.* Journal of Environmental Quality, 2009. **38**(3): p. 1053-1065.

94. Castlehouse, H., et al., *Biotransformation and accumulation of arsenic in soil amended with seaweed.* Environ Sci Technol, 2003. **37**(5): p. 951-7.

95. Antaya, N.T., et al., *Incremental amounts of Ascophyllum nodosum meal do not improve animal performance but do increase milk iodine output in early lactation dairy cows fed high-forage diets1.* Journal of Dairy Science, 2015. **98**(3): p. 1991-2004.

96. Bowell, R.J., *Sorption of Arsenic by Iron-Oxides and Oxyhydroxides in Soils.* Applied Geochemistry, 1994. **9**(3): p. 279-286.

97. Luxton, T.P., C.J. Tadanier, and M.J. Eick, *Mobilization of arsenite by competitive interaction with silicic acid.* Soil Science Society of America Journal, 2006. **70**(1): p. 204-214.

98. Meharg, A.A. and F.J. Zhao, *Arsenic & rice*. 2012, Dordrecht: Springer Verlag. xi, 171 p.

99. Wang, P., et al., *A review on completing arsenic biogeochemical cycle: Microbial volatilization of arsines in environment.* Journal of Environmental Sciences, 2014. **26**(2): p. 371-381.

100. Turpeinen, R., M. Pantsar-Kallio, and T. Kairesalo, *Role of microbes in controlling the speciation of arsenic and production of arsines in contaminated soils.* Science of The Total Environment, 2002. **285**(1–3): p. 133-145.

101. Pongratz, R., *Arsenic speciation in environmental samples of contaminated soil.* Science of the Total Environment, 1998. **224**(1-3): p. 133-141.

102. Frohne, T., et al., *Controlled variation of redox conditions in a floodplain soil: Impact on metal mobilization and biomethylation of arsenic and antimony.* Geoderma, 2011. **160**(3-4): p. 414-424.

103. Jia, Y., et al., *Microbial Arsenic Methylation in Soil and Rice Rhizosphere.* Environmental Science & Technology, 2013. **47**(7): p. 3141-3148.

104. Mestrot, A., et al., *Arsenic volatilization in model anaerobic biogas digesters.* Applied Geochemistry, 2013. **33**: p. 294-297.

105. Edvantoro, B.B., et al., *Microbial formation of volatile arsenic in cattle dip site soils contaminated with arsenic and DDT.* Applied Soil Ecology, 2004. **25**(3): p. 207-217.

106. Liu, S., et al., *Arsenic removal from contaminated soil via biovolatilization by genetically engineered bacteria under laboratory conditions.* Journal of Environmental Sciences, 2011. **23**(9): p. 1544-1550.

107. Yang, J.K., et al., *Adsorption, oxidation, and bioaccessibility of As(III) in soils.* Environmental Science & Technology, 2005. **39**(18): p. 7102-7110.

108. Newman, E.I., *The rhizosphere: carbon sources and microbial populations.* , in *Ecological Interactions in Soil: Plants, Microbes and Animals*, A.H. Fitter, D. Atkinson, and D.J. Read, Editors. 1985, Blackwell Scientific: Oxford, UK. p. 107-121.

109. Acosta, J.A., J.M. Arocena, and A. Faz, *Speciation of arsenic in bulk and rhizosphere soils from artisanal cooperative mines in Bolivia.* Chemosphere, 2015. **138**: p. 1014-1020.

110. Wu, C., et al., *The effect of silicon on iron plaque formation and arsenic accumulation in rice genotypes with different radial oxygen loss (ROL).* Environmental Pollution, 2016. **212**: p. 27-33.

111. Seyfferth, A.L., *Abiotic effects of dissolved oxyanions on iron plaque quantity and mineral composition in a simulated rhizosphere.* Plant and Soil, 2015. **397**(1-2): p. 43-61.

112. Yamaguchi, N., et al., *Arsenic Distribution and Speciation near Rice Roots Influenced by Iron Plaques and Redox Conditions of the Soil Matrix.* Environmental Science & Technology, 2014. **48**(3): p. 1549-1556.

113. Lee, C.-H., et al., *Iron plaque formation and its effect on arsenic uptake by different genotypes of paddy rice.* Plant and Soil, 2013. **363**(1-2): p. 231-241.

114. Hossain, M.B., et al., *The effects of iron plaque and phosphorus on yield and arsenic accumulation in rice.* Plant Soil, 2009. **317**(1-2): p. 167-176.

115. Wu, C., et al., *Do radial oxygen loss and external aeration affect iron plaque formation and arsenic accumulation and speciation in rice?* Journal of Experimental Botany, 2012. **63**(8): p. 2961-2970.

116. Li, R.Y., et al., *Uptake and Accumulation Characteristics of Arsenic and Iron Plaque in Rice at Different Growth Stages.* Communications in Soil Science and Plant Analysis, 2015. **46**(19): p. 2509-2522.
117. Mei, X.Q., et al., *The effects of radial oxygen loss on arsenic tolerance and uptake in rice and on its rhizosphere.* Environmental Pollution, 2012. **165**: p. 109-117.
118. Gadd, G.M., *Metals, minerals and microbes: geomicrobiology and bioremediation.* Microbiology-Sgm, 2010. **156**: p. 609-643.
119. Wang, Y., et al., *Release and transformation of arsenic from As-bearing iron minerals by Fe-reducing bacteria.* Chemical Engineering Journal, 2016. **295**: p. 29-38.
120. Newman, D.K., et al., *Dissimilatory arsenate and sulfate reduction in Desulfotomaculum auripigmentum sp. nov.* Archives of Microbiology, 1997. **168**(5): p. 380-388.
121. Macy, J.M., et al., *Two new arsenate/sulfate-reducing bacteria: mechanisms of arsenate reduction.* Archives of Microbiology, 2000. **173**(1): p. 49-57.
122. Wang, J., et al., *Biotransformation and biomethylation of arsenic by Shewanella oneidensis MR-1.* Chemosphere, 2016. **145**: p. 329-335.
123. Smeaton, C.M., et al., *Simultaneous Release of Fe and As during the Reductive Dissolution of Pb-As Jarosite by Shewanella putrefaciens CN32.* Environmental Science & Technology, 2012. **46**(23): p. 12823-12831.
124. Jia, Y., et al., *Microbial Arsenic Methylation in Soil and Rice Rhizosphere.* Environmental Science & Technology, 2013. **47**(7): p. 3141-3148.
125. Xu, L., et al., *Speciation change and redistribution of arsenic in soil under anaerobic microbial activities.* Journal of Hazardous Materials, 2016. **301**: p. 538-546.
126. Carey, A.M., et al., *Phloem transport of arsenic species from flag leaf to grain during grain filling.* New Phytologist, 2011. **192**(1): p. 87-98.
127. Carey, A.-M., et al., *A review of recent developments in the speciation and location of arsenic and selenium in rice grain.* Analytical and Bioanalytical Chemistry, 2011. **402**: p. 3275-3286.
128. Carey, A.M., et al., *Grain unloading of arsenic species in rice.* Plant Physiology, 2010. **152**(1): p. 309-319.

129. Baker, M.D., et al., *Methylation of Arsenic by Fresh-Water Green-Algae.* Canadian Journal of Fisheries and Aquatic Sciences, 1983. **40**(8): p. 1254-1257.
130. Hansen, H.R., et al., *Identification of tetramethylarsonium in rice grains with elevated arsenic content.* Journal of Environmental Monitoring, 2011. **13**(1): p. 32-34.
131. Arao, T., et al., *Effects of Arsenic Compound Amendment on Arsenic Speciation in Rice Grain.* Environmental Science & Technology, 2011. **45**(4): p. 1291-1297.
132. Andres, J. and P.N. Bertin, *The microbial genomics of arsenic.* Fems Microbiology Reviews, 2016. **40**(2): p. 299-322.
133. Zhang, S., et al., *Cometabolic biotransformation of fenpropathrin by Clostridium species strain ZP3.* Biodegradation, 2011. **22**(5): p. 869-75.
134. Stewart, E.J., *Growing Unculturable Bacteria.* Journal of Bacteriology, 2012. **194**(16): p. 4151-4160.
135. Xiao, K.-Q., et al., *Metagenomic analysis revealed highly diverse microbial arsenic metabolism genes in paddy soils with low-arsenic contents.* Environmental Pollution, 2016. **211**: p. 1-8.
136. Ma, L.Q., et al., *A fern that hyperaccumulates arsenic (vol 409, pg 579, 2001).* Nature, 2001. **411**(6836): p. 438-U3.
137. Sharma, I., *Arsenic induced oxidative stress in plants.* Biologia, 2012. **67**(3): p. 447-453.
138. Le, X.C., et al., *Determination of monomethylarsonous acid, a key arsenic methylation intermediate, in human urine.* Environmental Health Perspectives, 2000. **108**(11): p. 1015-1018.
139. Finnegan, P.M. and W. Chen, *Arsenic toxicity: the effects on plant metabolism.* Front Physiol, 2012. **3**: p. 182.
140. Deuel, L.E. and A.R. Swoboda, *Arsenic toxicity to cotton and soybeans.* Journal of Environmental Quality, 1972. **1**(3): p. 317-320.
141. Davis, R.D., P.H.T. Beckett, and E. Wollan, *Critical levels of twenty potentially toxic elements in young spring barley.* Plant Soil, 1978. **49**: p. 395-408.
142. Adriano, D.C., *Trace Elements in Terrestrial Environments*. 2nd ed. 2001, New York: Springer Verlag. 867.

143. Marin, A.R., P.H. Masscheleyn, and W.H. Patrick, *The influence of chemical form and concentration of arsenic on rice growth and tissue arsenic concentration.* Plant and Soil, 1992. **139**: p. 175-183.

144. Carbonell-Barrachina, A.A., et al., *The influence of arsenic chemical form and concentration on Spartina patens and Spartina alterniflora growth and tissue arsenic concentration.* Plant and Soil, 1998. **198**: p. 33-43.

145. Choudhury, B., S. Chowdhury, and A.K. Biswas, *Regulation of growth and metabolism in rice (Oryza sativa L.) by arsenic and its possible reversal by phosphate.* J. Plant Interact., 2011. **6**(1): p. 15-24.

146. Shri, M., et al., *Effect of arsenic on growth, oxidative stress, and antioxidant system in rice seedlings.* Ecotoxicol. Environ. Safety, 2009. **72**(4): p. 1102-1110.

147. Chakrabarty, D., et al., *Comparative transcriptome analysis of arsenate and arsenite stresses in rice seedlings.* Chemosphere, 2009. **74**(5): p. 688-702.

148. Ullrich-Eberius, C.I., A. Sanz, and A.J. Novacky, *Evaluation of arsenate-associated and vanadate-associated changes of electrical membrane potential and phosphate transport in Lemna gibba G1.* J. Exp. Bot., 1989. **40**(210): p. 119-128.

149. Raab, A., et al., *Uptake and translocation of inorganic and methylated arsenic species by plants.* Environmental Chemistry, 2007. **4**(3): p. 197-203.

150. Finnegan, P.M. and W.H. Chen, *Arsenic toxicity: the effects on plant metabolism.* Frontiers in Physiology, 2012. **3**.

151. Shin, H., et al., *Phosphate transport in Arabidopsis: Pht1;1 and Pht1;4 play a major role in phosphate acquisition from both low- and high-phosphate environments.* Plant J, 2004. **39**(4): p. 629-42.

152. Remy, E., et al., *The Pht1;9 and Pht1;8 transporters mediate inorganic phosphate acquisition by the Arabidopsis thaliana root during phosphorus starvation.* New Phytol, 2012. **195**(2): p. 356-71.

153. Jia, H., et al., *The phosphate transporter gene OsPht1;8 is involved in phosphate homeostasis in rice.* Plant Physiol, 2011. **156**(3): p. 1164-75.

154. Ye, Y., et al., *The Phosphate Transporter Gene OsPht1;4 Is Involved in Phosphate Homeostasis in Rice.* PLoS One, 2015. **10**(5): p. e0126186.

155. Sun, S., et al., *A constitutive expressed phosphate transporter, OsPht1;1, modulates phosphate uptake and translocation in phosphate-replete rice.* Plant Physiol, 2012. **159**(4): p. 1571-81.

156. Li, R.Y., et al., *The rice aquaporin Lsi1 mediates uptake of methylated arsenic species.* Plant Physiol, 2009. **150**(4): p. 2071-80.
157. Ma, J.F., et al., *Transporters of arsenite in rice and their role in arsenic accumulation in rice grain.* Proc Natl Acad Sci U S A, 2008. **105**(29): p. 9931-5.
158. Abedin, M.J., J. Feldmann, and A.A. Meharg, *Uptake kinetics of arsenic species in rice plants.* Plant Physiol, 2002. **128**(3): p. 1120-8.
159. Pommerrenig, B., T.A. Diehn, and G.P. Bienert, *Metalloido-porins: Essentiality of Nodulin 26-like intrinsic proteins in metalloid transport.* Plant Sci, 2015. **238**: p. 212-27.
160. Mitani, N., et al., *Identification and Characterization of Maize and Barley Lsi2-Like Silicon Efflux Transporters Reveals a Distinct Silicon Uptake System from That in Rice.* Plant Cell, 2009. **21**(7): p. 2133-2142.
161. Ma, J.F., et al., *An efflux transporter of silicon in rice.* Nature, 2007. **448**(7150): p. 209-12.
162. Zhao, F.J., et al., *The role of the rice aquaporin Lsi1 in arsenite efflux from roots.* New Phytol, 2010. **186**(2): p. 392-9.
163. Song, W.Y., et al., *A rice ABC transporter, OsABCC1, reduces arsenic accumulation in the grain.* Proc Natl Acad Sci U S A, 2014. **111**(44): p. 15699-704.
164. Schneider, S., et al., *Arabidopsis INOSITOL TRANSPORTER4 Mediates High-Affinity H(+) Symport of Myoinositol across the Plasma Membrane.* Plant Physiol, 2006. **141**(2): p. 565-77.
165. Schneider, S., et al., *Arabidopsis INOSITOL TRANSPORTER2 Mediates H+ Symport of Different Inositol Epimers and Derivatives across the Plasma Membrane.* Plant Physiology, 2007. **145**(4): p. 1395-1407.
166. Duan, G.-L., et al., *Inositol transporters AtINT2 and AtINT4 regulate arsenic accumulation in Arabidopsis seeds.* Nature Plants, 2015. **2**: p. 15202.
167. Zenk, M.H., *Heavy metal detoxification in higher plants--a review.* Gene, 1996. **179**(1): p. 21-30.
168. Schmoger, M.E., M. Oven, and E. Grill, *Detoxification of arsenic by phytochelatins in plants.* Plant Physiol, 2000. **122**(3): p. 793-801.
169. Sadee, B.A., M.E. Foulkes, and S.J. Hill, *A study of arsenic speciation in soil, irrigation water and plant tissue: A case study of the broad bean plant, Vicia faba.* Food Chemistry, 2016. **210**: p. 362-370.

170. Pizarro, I., et al., *Bioaccessibility and arsenic speciation in carrots, beets and quinoa from a contaminated area of Chile.* Science of The Total Environment, 2016. **565**: p. 557-563.
171. Zhao, F.J., et al., *Arsenic uptake and metabolism in plants.* New Phytologist, 2009. **181**: p. 777-794.
172. Ma, J.F., et al., *An efflux transporter of silicon in rice.* Nature, 2007. **448**: p. 209-12.
173. Seyfferth, A., et al., *Defining the distribution of arsenic species and plant nutrients in rice (Oryza sativa L.) from the root to the grain.* Geochim. Cosmochim. Acta, 2011. **75**: p. 6655-6671.
174. Williams, P.N., et al., *Variation in arsenic speciation and concentration in paddy rice related to dietary exposure.* Environ. Sci. Technol., 2005. **39**(15): p. 5531-5540.
175. Hughes, M.F., *Arsenic toxicity and potential mechanisms of action* Toxicology Letters, 2002. **133**(1): p. 1-16.
176. Sanchez-Bermejo, E., et al., *Natural variation in arsenate tolerance identifies an arsenate reductase in Arabidopsis thaliana.* Nat Commun, 2014. **5**: p. 4617.
177. Chao, D.Y., et al., *Genome-wide association mapping identifies a new arsenate reductase enzyme critical for limiting arsenic accumulation in plants.* PLoS Biol, 2014. **12**(12): p. e1002009.
178. Styblo, M., et al., *Comparative toxicity of trivalent and pentavalent inorganic and methylated arsenicals in rat and human cells.* Archives of Toxicology, 2000. **74**(6): p. 289-299.
179. Marin, A.R., P.H. Masscheleyn, and W.H. Patrick, *The Influence of Chemical Form and Concentration of Arsenic on Rice Growth and Tissue Arsenic Concentration.* Plant and Soil, 1992. **139**(2): p. 175-183.
180. Rosen, B.P., *Families of arsenic transporters.* Trends Microbiol, 1999. **7**(5): p. 207-12.
181. Song, W.Y., et al., *A rice ABC transporter, OsABCC1, reduces arsenic accumulation in the grain.* Proc. Natl. Sci. U.S.A., 2014. **111**(44): p. 15699-704.
182. Song, W.Y., et al., *Arsenic tolerance in Arabidopsis is mediated by two ABCC-type phytochelatin transporters.* Proc Natl Acad Sci U S A, 2010. **107**(49): p. 21187-92.

183. Hartley-Whitaker, J., G. Ainsworth, and A.A. Meharg, *Copper- and arsenate-induced oxidative stress in Holcus lanatus L. clones with differential sensitivity.* Plant Cell Environ., 2001. **24**(7): p. 713-722.

184. Lee, K.W., et al., *Overexpression of alfalfa mitochondrial HSP23 in prokaryotic and eukaryotic model systems confers enhanced tolerance to salinity and arsenic stress.* Biotechnol. Lett., 2012. **34**(1): p. 167-174.

185. Mascher, R., et al., *Arsenate toxicity: effects on oxidative stress response molecules and enzymes in red clover plants.* Plant Sci., 2002. **163**(5): p. 961-969.

186. Stoeva, N., M. Berova, and Z. Zlatev, *Physiological response of maize to arsenic contamination.* Biologia Plant., 2003. **47**(3): p. 449-452.

187. Dixit, S. and J.G. Hering, *Comparison of arsenic(V) and arsenic(III) sorption onto iron oxide minerals: Implications for arsenic mobility.* Environmental Science & Technology, 2003. **37**(18): p. 4182-4189.

188. Li, R.Y., et al., *Mitigation of Arsenic Accumulation in Rice with Water Management and Silicon Fertilization.* Environmental Science & Technology, 2009. **43**(10): p. 3778-3783.

189. Grassi, C., et al., *Arobic rice: crop performance and water use efficiency.* Journal of Agriculture and Environment for International Development, 2009. **103**(4): p. 259-270.

190. Arao, T., et al., *Effects of Water Management on Cadmium and Arsenic Accumulation and Dimethylarsinic Acid Concentrations in Japanese Rice.* Environmental Science & Technology, 2009. **43**(24): p. 9361-9367.

191. Hu, P.J., et al., *Water management affects arsenic and cadmium accumulation in different rice cultivars.* Environmental Geochemistry and Health, 2013. **35**(6): p. 767-778.

192. Hu, P.J., et al., *Effect of water management on cadmium and arsenic accumulation by rice (Oryza sativa L.) with different metal accumulation capacities.* Journal of Soils and Sediments, 2013. **13**(5): p. 916-924.

193. Honma, T., et al., *Effects of soil amendments on arsenic and cadmium uptake by rice plants (Oryza sativa L. cv. Koshihikari) under different water management practices.* Soil Science and Plant Nutrition, 2016. **62**(4): p. 349-356.

194. Matsumoto, S., et al., *Evaluation of the effects of application of iron materials on the accumulation and speciation of arsenic in rice grain grown on uncontaminated soil with relatively high levels of arsenic.* Environmental and Experimental Botany, 2016. **125**: p. 42-51.

195. Farrow, E.M., et al., *Reducing arsenic accumulation in rice grain through iron oxide amendment.* Ecotoxicology and Environmental Safety, 2015. **118**: p. 55-61.

196. Chou, M.L., et al., *Inhibition of ethylenediaminetetraacetic acid ferric sodium salt (EDTA-Fe) and calciumperoxide (CaO2) on arsenic uptake by vegetables in arsenic-rich agricultural soil.* Journal of Geochemical Exploration, 2016. **163**: p. 19-27.

197. Tiberg, C., et al., *Immobilization of Cu and As in two contaminated soils with zero-valent iron - Long-term performance and mechanisms.* Applied Geochemistry, 2016. **67**: p. 144-152.

198. Penido, E.S., et al., *Biogeochemical impacts of silicon-rich rice residue incorporation into flooded soils: Implications for rice nutrition and cycling of arsenic.* Plant and Soil, 2016. **399**(1-2): p. 75-87.

199. Meharg, C. and A.A. Meharg, *Silicon, the silver bullet for mitigating biotic and abiotic stress, and improving grain quality, in rice?* Environmental and Experimental Botany, 2015. **120**: p. 8-17.

200. Seyfferth, A.L., et al., *Soil Incorporation of Silica-Rich Rice Husk Decreases Inorganic Arsenic in Rice Grain.* Journal of Agricultural and Food Chemistry, 2016. **64**(19): p. 3760-3766.

201. Norton, G.J., et al., *Identification of Low Inorganic and Total Grain Arsenic Rice Cultivars from Bangladesh.* Environmental Science & Technology, 2009. **43**(15): p. 6070-6075.

202. Duan, G., et al., *Expressing ScACR3 in rice enhanced arsenite efflux and reduced arsenic accumulation in rice grains.* Plant Cell Physiol, 2012. **53**(1): p. 154-63.

203. Song, W.Y., et al., *Arsenic tolerance in Arabidopsis is mediated by two ABCC-type phytochelatin transporters.* Proc. Natl. Acad. Sci. U.S.A., 2010. **107**: p. 21187-92.

204. Norton, G.J., et al., *Genome wide association mapping of grain arsenic, copper, molybdenum and zinc in rice (Oryza sativa L.) grown at four international field sites.* PLoS One, 2014. **9**(2): p. e89685.

205. Tang, Z., et al., *Arsenic Methylation in Arabidopsis thaliana Expressing an Algal Arsenite Methyltransferase Gene Increases Arsenic Phytotoxicity.* J Agric Food Chem, 2016. **64**(13): p. 2674-81.

Understanding arsenic dynamics in agronomic systems to predict and prevent uptake by crop plants.

Tracy Punshon*[1], Brian P. Jackson[2], Andrew A. Meharg[3], Todd Warczack[1], Kirk Scheckel[4] and Mary Lou Guerinot[1].

*Corresponding author:

[1]Dartmouth College, Department of Biology, 78 College Street, Hanover, New Hampshire, 03755, USA. tracy.punshon@dartmouth.edu, mary.lou.guerinot@dartmouth.edu, Todd.M.Warczack.GR@dartmouth.edu

[2]Dartmouth College, Department of Earth Sciences, Hanover, New Hampshire, 03755, USA. bpj@dartmouth.edu

[2]Institute for Global Food Security, Queen's University Belfast, Belfast BT9 5HN, United Kingdom. aa.meharg@qub.ac.uk

[4]USEPA Office of Research and Development, National Risk Management Laboratory, 26 West Martin Luther King Drive, Cincinnati, OH 45224. scheckel.kirk@epa.gov

Keywords: arsenic, sources, soil, agriculture, plants, mitigation

Abstract

This review is on arsenic in agronomic systems, and covers processes that influence the entry of arsenic into the human food supply. The scope is from sources of arsenic (natural and anthropogenic) in soils, biogeochemical and rhizosphere processes that control arsenic speciation and availability, through to mechanisms of uptake by crop plants and potential mitigation strategies. This review makes a case for taking steps to prevent or limit crop uptake of arsenic, wherever possible, and to work toward a long-term solution to the presence of arsenic in agronomic systems. The past two decades have seen important advances in our understanding of how biogeochemical and physiological processes influence human exposure to soil arsenic, and this must now prompt an informed reconsideration and unification of regulations to protect the quality of agricultural and residential soils.

1. Introduction

Consumption of staple foods such as rice, beverages such as apple juice, or vegetables grown in historically arsenic-contaminated soils are now recognized as tangible routes of arsenic exposure. The presence of elevated concentrations of arsenic in the soil is not a pre-requisite for dietary arsenic exposure; seen in the accumulation of arsenic by rice grown in uncontaminated soils[1]. When drinking-water arsenic concentrations are low, dietary arsenic can be a significant exposure[2]. Understanding the sources of arsenic to crop plants and the factors that influence them is key to reducing human exposure now and preventing exposure in future. In addition to the abundant natural sources of arsenic, there are a large number of industrial and agricultural sources of arsenic to the soil; from mining wastes, coal fly ash, glass manufacturing, pesticide application, wastewater sludge, pharmaceutical waste, livestock dips, smelting activities to phosphate fertilizers. Plant uptake of arsenic was previously assumed to be too low to merit setting limits for arsenic in food crops, but given that measurable biological effects occur in at arsenic levels below the current maximum contaminant level (MCL) for drinking water[3], these low levels can still translate into significant exposures, particularly in children[4] and presumably in adults who consume a lot of rice. In response, the World Health Organization (WHO) set an advisory MCL for inorganic arsenic in white (polished) rice of 0.2 mg/kg[5] along with the limit of 10 μg/L in water, and the European Union set similar standards that included a lower MCL (0.1 mg/kg) for rice-containing baby foods[6]. Currently, dietary arsenic exposure is suspected to play a role in cardiovascular disease in adults[7], and to disrupt the glucocorticoid system (involved in learning and memory) to those exposed *in utero*[8]. An in depth review of the current findings on the relationship between dietary arsenic exposure and human health is provided by Davis et al. (this issue).

In the United States, regulations on arsenic are distributed to several agencies. The Environmental Protection Agency (EPA) developed the MCL for arsenic in drinking water (10 μg/L) in 2006; a level supported by the World Health Organization, Canada and the European Union. In the state of New Jersey (USA) the limit is 5 μg/L, and in Australia, 7 μg/L. Many other nations still adopt a level of 50 μg/L (Bahrain, Bangladesh, Bolivia, China, Egypt, India, Indonesia, Oman, Philippines, Saudi Arabia, Sri Lanka, Vietnam, Zimbabwe)[9], with the exception of Mexico (35 μg/L). In the USA, The Food and Drug Administration (FDA) is responsible for setting action levels for arsenic in food, which includes apple and pear juice at 10 μg/L, in line with EPA's drinking water MCL. In Canada, the Canadian Food Inspection Agency issued alerts on excessive arsenic in rice and pear products in 2014. Consistent with the European Commission's limit for arsenic in rice used in food production for infants and young children, the FDA is proposing an action level of 0.1 mg/kg for inorganic arsenic in infant rice cereal[10]. Foods in Australia and New Zealand may not contain more than 1 mg/kg dry mass of arsenic, and salt for food use must not contain more than 0.5 mg/kg. Japan has a limit of 15 mg/kg of arsenic in paddy soils[11]. Likewise, Thailand has an agricultural arsenic soil quality standard of 3.9 mg/kg. Within the USA, states differ widely in their action levels for arsenic in soil, for instance New Jersey has a cleanup criterion of 20 mg/kg and Florida has a cleanup target level of 2.1 mg/kg and 12 mg/kg for industrial sites[12].

Arsenic occurs in food because it is present in soil and water and is taken up by plants. This review article brings together the latest scientific information on arsenic in agronomic systems, describing its sources in soils and the processes that influence the uptake of arsenic by crop plants. The intention of this review is to prompt a reconsideration and unification of government regulations on action levels for arsenic in agricultural soil; raise awareness of how both former and ongoing inputs of arsenic to soil can result in food contamination and impacts to human health and finally, to indicate the way forward for mitigation strategies that safeguard valuable soil resources.

2. Natural sources of arsenic in soil

Below toxic concentrations, the higher the total soil arsenic concentration (the sum of all arsenic species, regardless of bioavailability) the higher the crop uptake of arsenic. This is true of anaerobic cultivation systems such as rice[13-15], aerobic horticultural systems[16] as well as conventional (aerobic) agriculture[15]. The global average total soil arsenic concentration is 5 mg/kg, (equivalent to parts per million), but there is large variation between and within geographical regions[17]. Where soils have formed on arsenic-rich bedrock, or downstream of these bedrocks, very high concentrations of natural arsenic can result. Concentrations of up to 4000 mg/kg arsenic have been measured in soils from the arsenopyrite belt (iron arsenic sulphide, FeAsS) in Styria, Austria[18], for instance. There are approximately 568 known minerals that contain arsenic as a critical component[19]. Arsenic is present in many rock-forming minerals because it can chemically substitute for phosphorus (V), silicate (IV), aluminum (III), iron (III) and titanium (IV) in mineral structures. Global mapping data of total arsenic concentrations in topsoil is not available, although large-scale regional maps are available for soil arsenic concentrations in Europe[20] and the USA [21]. European data predicts that most soils range < 7.5 - 20 mg/kg arsenic, with a median of 6 mg/kg[20]. This prediction comes from block regression-kriging; a spatial prediction technique based on regressing soil arsenic concentrations against auxiliary variables, and is useful because it uses a particularly high resolution (block size of 5 km^2). On a continental scale, large zones of soils with approximately 30 mg/kg arsenic have been found in southern France, the north-eastern Iberian Peninsula and south-west England, with the two latter being zones of extensive natural mineralization associated with base and precious metal mining activities. The United State Geological Survey (USGS) soil sampling of the contiguous USA reports a mean soil arsenic concentration of approximately 5 mg/kg with 5 and 95 percentile values of approximately 1.3 and 13 mg/kg respectively[22]. Large regional patterns are apparent in the data, for example the soils of New Hampshire have soil arsenic concentrations of approximately 10 mg/kg arsenic, and Florida, 3.5 mg/kg. The sampling density goal for the USA surface soils and stream sediments database is 1 per 289 km^2 [23], but is currently at only 1 sample per 1600 km^2. This contrasts with smaller regional surveys such as the recently published Tellus database for Northern Ireland that has a sampling density of 2 km^2 [24] (median total soil arsenic concentration 8.7 mg/kg). At this sampling density, fine-scale data for factors shown to affect soil arsenic, such as bedrock type, altitude and organic matter for instance, can be observed, providing the opportunity to make predictions about arsenic bioavailability and mobility.

Soil or sediment arsenic concentrations are the result of the complex and dynamic interplay between inputs and outputs[25]. Natural sources of arsenic to agronomic catchments are dominated by bedrock weathering (mechanical, chemical and biological) and depositional inputs, with the ultimate sinks at the base of catchments often being a significant distance from sources[26]. Outputs include leaching into water bodies (vertically and horizontally), soil erosion[25] and biovolatilization[27]. In arid regions surface evaporation of water can lead to arsenic enrichment from the draw up of subsurface water[25] and from waters used in crop irrigation[28]. Mass-balances (accounting for all inputs and outputs for a particular ecosystem) are rarely conducted for arsenic fluxes within catchment areas, but a good example is from a mining-impacted catchment area[29], where chemical weathering, followed by mechanical weathering dominated arsenic inputs, which were primarily from arsenopyrite. Similarly, in a gold-mining region, weathering contributed an estimated 95% of the arsenic[30]. In a forested catchment area, where atmospheric arsenic inputs were the dominant source to highly organic soil (soils with more than 10% organic matter), inputs of arsenic via precipitation were ~6 g/ha/y[31], and organic soils were a net source of arsenic, while mineral soils (less than 10% organic matter) were a sink. This agrees with depositional inputs of arsenic measured in the UK, which ranged from ~1 to ~10 g arsenic/ha/y[32]. UK regional scale maps show that arsenic deposition is highest at altitude and in the west of the country; the least polluted regions with air masses originating in the Atlantic. This suggests a marine source of arsenic. Depositional maps relate well to soil arsenic maps such as in maps of Northern Ireland and England[33] that show highest arsenic concentrations in peat soils at higher altitude, along with bedrock geological anomalies. Peat soils at higher altitude are sinks for arsenic, and become sources if the peat is mineralized or eroded. The topic of upland organic soils acting as sinks and sources of arsenic is receiving more research attention[34], and could be important on a regional scale as a source of arsenic to downstream sediments.

In large catchment areas of continental importance, such as the deltas that form to the south and east of the Himalayas, plate tectonic-derived mechanical weathering is thought to be the most important source of arsenic. One theory is that the mechanical weathering caused by Pleistocene tectonic uplift in the Himalayas is the key to understanding why arsenic is so elevated in Holocene aquifers, such as those of SE Asia, and in the glacial tills of Europe and North America[26]. Mechanical weathering of bedrock exposes previously inaccessible mineral surfaces, and the finer grinding leads to enhanced surface areas for chemical and microbial weathering to take place, causing greater solubilisation of arsenic[25, 26, 35]. Chemical and microbial weathering can take place at or near the source, or in sediment sinks. For instance, bacteria isolated from Bay of Bengal aquifers can mobilize arsenic from apatite[35] (See Section 5). Invariably, the arsenic loadings into soil will be dependent on arsenic in the bedrock, and the extent of the weathering of that bedrock-derived material along the route from source to sink. Soils with basalt bedrock had the lowest median arsenic content, while those with psammite, semipelite, and lithic arsenite bedrocks had the highest. Interpretation of such fine-scale mapping can ultimately lead to predictions of soil arsenic concentrations where detailed maps are not available. Combined with an understanding of soil chemistry, this will enhance the ability to predict elevated concentrations of arsenic in crops[36].

3. Anthropogenic sources of arsenic to soil

Many anthropogenic activities have increased soil arsenic concentrations above the natural, background levels mentioned in Section 2 above, and they have the potential to increase the arsenic concentration in food. This is especially the case in the USA where the widespread use of arsenic-based herbicides, pesticides and livestock antibiotics throughout the 20th century has ultimately increased the arsenic concentrations of current productive USA agricultural soils[37-39].

3.1. Base and precious metal mining

The dominant mineral source of arsenic is thought to be pyrite (iron sulfide, FeS_2)[40], an economically important ore deposit. High arsenic concentrations are found in many oxide minerals and hydrous metal oxides, either part of their structure or as sorbed and occluded species[41]. Iron oxides accumulate arsenic up to concentrations of several weight percent (1 weight % being equivalent to 10,000 mg/kg), and arsenic tends to bind to iron (III) (hydr)oxides whenever they are present. Arsenic is found predominantly as arsenopyrite but also can occur as orpiment (arsenic trisulphide As_2S_3), realgar (α-As_4S_4) and other arsenic sulfide minerals[42, 43]. Arsenic is a byproduct of most mining operations and is present at high concentrations in the mine waste, and, because arsenic sulfides are particularly prone to oxidation in surface environments, in mining wastewaters[42, 44]. Arsenic can constitute 1% or more of the ore and solid waste, and wastewaters and impacted streams often contain dissolved arsenic concentrations ranging from 0.01 to over 10 mg/L. Because mining and smelting operations are localized, arsenic contamination of soils exists around the mine site with the concentration decreasing with distance from the source. Windblown dispersion of fine particulate material is a particular problem, spreading contamination greater distances from the mine site. This fine material - which is not completely removed by washing[16] - can directly contaminate plant material; especially leafy material with high surface area. This presents a tangible risk to residents and home gardeners in the vicinity of areas with significant surface soil arsenic contamination. A comparison of arsenic concentrations in vegetables grown in SW England (the site of historic mining activities) with those from a pristine site in NW Scotland found a generally good correlation between total plant arsenic and soil arsenic concentrations. Increased arsenic concentrations were measured in produce from SW England where soil arsenic concentrations ranged from 120 – 1130 mg/kg. Arsenic concentrations were high in leafy greens (kale, spinach, lettuce) and some unpeeled vegetables (potatoes, swedes, carrots) were higher than when peeled, which, in both cases, points to contamination from windblown soil particles and soil adhesion to below ground biomass, rather than from root uptake. In this particular study, the majority of arsenic was present as the inorganic form[16]. Similar results were obtained from home gardens near the Iron King Mine Superfund Site in Arizona, USA[45, 46]. Here the tailings had arsenic concentrations of 3,710 mg/kg and residential soil sampled adjacent to the site ranged from 120 – 633 mg/kg. Edible plant tissue concentrations ranged from < 0.01 – 1.96 mg/kg (plant concentrations are expressed as dry weight throughout), and were generally positively correlated with soil arsenic concentrations. Leafy and high surface area vegetables such as lettuce, kale, broccoli and cabbage accumulated higher arsenic concentrations than beans, tomatoes, cucumbers and peppers. Arsenic in mine-affected vineyard soils in Italy ranged from 4 – 283 mg/kg and positive correlations were observed between soil concentrations and arsenic levels in vine leaves and grapes, however, levels in wine were low (< 1.62

μg/L)[47]. In the Hunan province, China, the high levels of inorganic arsenic in rice have been traced to mining activities in the area[48-50].

3.2. Coal combustion for energy

The concentration of arsenic in USA coal ranges from 1 – 71 mg/kg with an average concentration of 24 mg/kg[51]. Fly ash, the major byproduct of the coal combustion process, consists of fine particles that are driven out with the flue gases, and is a major source of arsenic to the wider environment. Coal ash is one of the most abundant of industrial wastes; close to 130 million tons[52] of coal fly was generated in the USA in 2014, with 100 million tons estimated from the European Union in 2011[53]. Arsenic concentrates in the fly ash during combustion of coal for energy; the median arsenic concentration in USA fly ash is 71 mg/kg[54]. Fly ash is often sluiced into settling basins, and because arsenic in fresh ash is quite soluble, wastewater arsenic concentrations can consequently be quite high. Arsenic can build up in the sediments of coal fly ash settling basins and reach concentrations of over 1000 mg/kg. Catastrophic failures of these setting basins have caused severe environmental problems and contaminated surface waters with arsenic[55]. There is a well-founded concern that arsenic from coal combustion wastes can contaminate soil and enter the food supply. The use of coal fly ash as a soil amendment can lead to elevated arsenic concentrations in crops (as well as boron, selenium and molybdenum), although its lack of soil macronutrients and the potential for arsenic toxicity prevents the sole application of coal fly ash as a soil amendment[56, 57]. Formulating ash/organic waste mixtures that conform to USEPA regulations for total arsenic application and meet soil and plant fertility requirements has been shown to safe and effective for agronomic use[58].

3.3. Pesticides

Perhaps the largest anthropogenic input of arsenic to agricultural soils in the USA is from the agricultural use of arsenic-based pesticides and herbicides for most of the 20th century. Calcium arsenate and lead arsenate were used extensively up to the 1950s, mostly on orchard soils to combat the codling moth. At peak, 132,000 metric tons of each pesticide compound was applied annually between 1930-1940[37]. In addition to apples, inorganic arsenic pesticides were used on a range of crops including essentially all fruit trees, vine berries, sweet potatoes, white potatoes, most vegetables and cotton[37]. Both lead and arsenate have long residence times in soils and high concentrations (often >100 mg/kg) of these two elements have been reported in old orchard soils in Washington[59], North Carolina[60], New Hampshire[61], New Jersey[62] and Virginia[63]. There is some evidence of greater mobility for arsenic (than lead)[61, 64], and retention of both elements depends on soil type and other environmental factors but most of this legacy contaminant remains in the soil[62]. Use of lead arsenate decreased after 1950s and was finally banned in 1988. The organic arsenic compounds dimethylarsinic acid (DMA) and monomethylarsonic acid (MMA) were used as pesticides on cotton and herbicides for golf courses and right-of-ways until they too were withdrawn from use in 2013. High levels of MMA were reported in transient surface waters adjacent to a crop sprayer operation[65]. Legacy soil arsenic contamination resulting from organic arsenical pesticides plays a major role in straighthead disease of rice[66] (See Section 6.1). It may be that arsenical pesticides have leached to groundwater, as has been suggested for the Texas High Plains Aquifer[67], although a study of the Ogallala aquifer in the High Plains in Texas found no evidence of anthropogenic arsenic in the groundwater[68]. Similarly, there was no relationship between groundwater arsenic and past (inorganic) arsenic pesticide usage in a comprehensive study of New Hampshire groundwater sources[69]. About 10%, depending on soil substrate, of monosodium methyl arsenate applied to sandy soils (simulated golf course greens) leached into percolating water. Demethylation and methylation occurred because both inorganic arsenic species and DMA were also detected in the percolating water[70]. As with mining-impacted soils, plants grown on soils that are high in arsenic from arsenical pesticide contamination take up higher levels of arsenic into their edible tissues, observed for example in potatoes[71], carrots[72] and leafy green vegetables[73, 74].

Former pesticide application has been suggested be a factor in the presence of higher levels of total arsenic found in rice grown in the south-central regions of the USA[75, 76] compared to other areas of the USA and to other countries, such as Bangladesh[77]. Evidence on varietal differences in arsenic uptake, speciation and distribution within rice grain (See also Section 6) strongly suggest that soil arsenic concentration is not the sole, nor particularly the main driver of this phenomenon. Factors likely to be influential include the differences in the soil microbial community composition between geographical regions that affect arsenic methylation, considering that plants themselves cannot methylate arsenic[78].

3.4. Wood preservatives

Chromated copper arsenate (CCA) is used as a wood preservative and was extensively used on decking and other residential usages until a voluntary manufacturer withdrawal in 2003. The primary health concern is for young children in direct contact with CCA-treated wood, but localized leaching of arsenic (as well as chromium and copper) also occurs to surrounding soil. Soil arsenic concentrations of 37 – 250 mg/kg have been reported for soils sampled near CCA-treated utility poles (N=12)[79] and mean arsenic concentrations for soils collected below decks and footbridges in Florida, USA was reported to be 28.5 mg/kg compared with a control concentration of 1.3 mg/kg (N= 65)[80]. Arsenic from CCA contaminated soils appears to be more bioavailable than from other anthropogenic sources to soil[81].

3.4. Organic manures

Land application of sewage sludge (biosolids) in the USA is regulated by Environmental Protection Agency Part 503 Biosolids rule; which set the maximum arsenic concentration of the sludge at 75 mg/kg, an annual pollutant-loading rate of 2.0 kilograms arsenic/hectare (kg/ha) and a cumulative pollution-loading rate of 41 kg/ha over the lifetime of applications. Assuming a plow layer of 17 cm, application at the maximum annual rate implies an approximate 1.2 mg/kg maximum increase in soil arsenic, while the cumulative maximum loading rate could increase soil arsenic concentrations by approximately 24 mg/kg over the lifetime of application and assuming no loss from the soil profile. This cumulative loading rate of 24 mg/kg is significant when considered against an average soil arsenic concentration of 5 mg/kg (See Section 2), however, relative to mine-impacted or inorganic arsenic pesticide impacted soils where arsenic concentrations are frequently > 100 mg/kg, it is of lesser concern. Also, sewage sludge is often high in aluminum and iron oxide phases, used in the flocculation process, which are efficient scavengers of inorganic arsenic thus lowering the arsenic bioavailability [82].

Arsenic occurs in animal wastes primarily because of the former use of arsenic antibiotics in poultry and turkey feed; until 2015 four drugs, roxarsone, p-arsanilic acid, carbarsone and nitrosone, were regulated for use, with roxarsone being the most prevalent. As of 2016 all four of these compounds have all been withdrawn from use[83]. All four are organic arsenic compounds with an arsenate functional group attached to a benzene ring, and differ by other substituents on the ring. The compounds are not readily adsorbed or metabolized and so occur at concentrations up to 40 mg/kg in animal manures. This provides three points of entry to the human food chain; directly through arsenic in chicken and turkey meat[38, 39, 84], from plant uptake after land application of manure, and runoff to surface water or groundwater. A number of studies have shown that these organic arsenic compounds can be degraded by both photolytic[85] and microbial[86] processes and that this degradation happens both during composting of stockpiled litter[87, 88] and after land application[89, 90]. Long term application of poultry litter to Upper Coastal Plain soils increased soil arsenic concentrations from 2.7 to 8.4 mg/kg after 25 years of application[91]. Similar increases have been reported for other southern states of the USA[92]. There is evidence to suggest that other soluble constituents of the litter, for example phosphate and dissolved organic carbon compounds, facilitate arsenic solubility and leaching[89, 92, 93].

3.5. Seaweed fertilizers

Seaweeds can contain far higher concentrations of arsenic than crop plants: up to 100 mg/kg (Taylor et al, this issue). In most cases the arsenic is present as arsenosugars, which are of low toxicity to humans (Taylor et al, this issue). However, as in the case of poultry litter, these compounds degrade (ultimately) to inorganic arsenic after land application[94]. Although seaweeds are a 'niche' soil amendment, their use agriculture is increasing and has been adopted by many organic farms as a soil fertilizer as well as a feed additive in organic dairy farming[95].

4. Biogeochemical cycling within terrestrial agronomic ecosystems

Arsenic cycles within the soil surface and near-surface environment[96], influenced by mineralogy, abiotic factors such as pH and redox potential (E_H), and biotic factors such as microbially-mediated biomethylation.

4.1. Redox regulation

The most important biogeochemical step in the exposure of humans to arsenic is its release from soils and sediments into pore water; the water contained within soil pores and/or rock[40]. With the exception of extreme pH conditions (<4 or >9), or high concentrations of competing ions (e.g. phosphate, silicic acid or silicate[97]) the release of arsenic from its strong bonds with soil particles depends upon redox potential (E_H); the extent of aeration of the soil[40]. As E_H falls, electron acceptors are depleted and anoxic conditions develop, causing iron oxides and oxyhydroxides to be reduced and dissolve, releasing sorbed arsenic into the soil solution[98] where it can be taken up by plant roots, or leached into groundwater.

Agronomic cropping systems can be divided with respect to arsenic mobilization on the basis of their redox status. Dominant biogeochemical processes influencing aerobic systems, specifically cereals, upland rice, fruit tree orchards, and community gardens, differ from those that dominate in anaerobic systems, predominantly in flooded rice paddies. In aerobic soils, arsenic speciation is predominantly arsenate (arsenic (V)), and is tightly bound to soil particles. Under anaerobic or flooded conditions, arsenic is reduced, and arsenite (arsenic (III)) is the dominant species[40]. Arsenite is less stably bound to aluminum hydroxides and aluminosilicate clay minerals in the soil than arsenate, for which they exhibit a much stronger binding preference[40]. With few exceptions (such as under conditions of sulfur release), transition of arsenic speciation from arsenate to arsenite is the most influential factor to arsenic bioavailability; and it is under anaerobic conditions where arsenic becomes an imminent human health concern. Influential biogeochemical processes in aerobic systems are ageing and accumulation of arsenic in soil, and in anaerobic systems reductive dissolution of iron-bearing minerals is the dominant process.

4.2. Biotransformation to methylated and volatile species

Volatile arsenicals are arsenic species with a boiling point below 150˚C; the most volatile of which is arsine gas (AsH_3), followed by monomethylarsine ($MeAsH_2$), dimethylarsine (Me_2AsH_2) and finally completely methylated trimethylarsine (TMA). Volatile arsenic species can be formed either biotically – by fungi, bacteria and algae[27, 99, 100]or abiotically[99]. In natural systems arsines readily react with oxygen to form non-volatile oxidation products, with AsH_3 most rapidly oxidized and challenging to detect in environmental samples. Oxidation of the arsine gases to inorganic arsenic species completes the arsenic cycle, with arsenic returned to the soil by rain or dry deposition[101].

Arsenic methylation in soils increases with decreasing redox potential[102], and addition of organic matter. Increased arsenic volatilization was measured in soil after the addition of rice straw[103], and animal waste products[104]. Inoculation of fungi (*Penicillium* and *Ulocladium* spp.) increased arsenic volatilization up to 8 fold in heavily contaminated and spiked soils[105]. Microbially mediated arsenic volatilization remains very inefficient, which hinders attempts to use it in soil remediation. Gaseous arsines are volatilized from arsenic contaminated soils into the atmosphere at very low rates: a microcosm study found 0.5 – 70 µg of arsenic kg^{-1} soil $year^{-1}$ was volatilized from a range of soils and a range of arsenic levels[27], and field measurements of arsenic volatilization are 1-2 orders of magnitude lower than those made in laboratory mesocosms [98].Genetic transformation of bacteria, using genes encoding for the protein product arsenite S-adenosyl methyltransferase (*ars*M) is an attempt to enhance arsenic methylation and volatilization. The *ars*M from *Rhodopseudomonas palustris* was expressed in *Sphingomonas desiccabilis* and

Bacillus idriensis grown in an aqueous system, resulting in a 10-fold increase in arsenic volatilization compared to the wild type strains. In a soil-based system, 2.2 – 4.5% of arsenic was removed via microbially-mediated volatilization over an incubation period of 30 days[106] (See also section 5.3).

4.3. Changes in soil arsenic bioavailability due to ageing

Although arsenic in aerobic soils has a lower bioavailability and presents less of an immediate concern for crop uptake, aerobic soil can accumulate arsenic from human inputs, retain them for long periods of time, and release them when redox conditions change (See Section 3.3). Human inputs of arsenic, as discussed in Section 3, are diverse; biosolids, sewage sludge, coal fly ash, poultry litter, industrial waste, arsenical pesticides and from irrigation with naturally arsenic-enriched groundwater. For aerobic soils, ageing – where binding stability of arsenic to soil particles increases over time, is a particularly important part of arsenic cycling. Factors controlling ageing of arsenic include soil type, organic matter content and arsenic species. Both inorganic and organic arsenic species are subject to ageing, with studies indicating a slow oxidation process from arsenite to arsenate over time[107].

5. Rhizosphere processes

Processes occurring in the rhizosphere (the boundary layer of soil under the influence of plant roots) dramatically influence arsenic concentrations and bioavailability because they involve local alterations in redox potential, pH and organic matter content. Rhizosphere acidification occurs during iron uptake by all plant species during cation uptake and charge balance, when protons are released into the rhizosphere. Plants release anywhere from 10 to 250 mg of carbon per gram of root tissue into the rhizosphere; about 10-40% of their total photosynthetically fixed carbon[108], making the rhizosphere particularly rich in organic carbon compared to bulk soil, which in turn exerts an influence on arsenic solubility by stimulating microbially-mediated reductive dissolution of soil minerals. Large differences have been found in the arsenic concentration of rhizosphere soils compared with bulk soils in highly arsenic-contaminated areas, with higher concentrations of arsenic in rhizosphere soils compared to bulk soils[109].

In anaerobic soils, the iron plaque that develops on the submerged stem and roots of rice plants dominates rhizosphere dynamics of arsenic. In flooded environments such as paddy fields, plants oxygenate the rhizosphere through specialized tissues called aerenchyma, which are found in many aquatic plants and emergent macrophysics such as rice. This radial oxygen loss creates an oxidized layer around plant tissue that stimulates aerobic microbial activity and the oxidation of iron, which precipitates and forms a visible iron plaque on the root surface [110-114]. Formation of an iron (oxyhydr)oxide plaque on root surfaces can alter the uptake of arsenic by rice, acting as a sorbent for excess nutrients such as ferrous iron (reduced iron) as well as arsenic and aluminum[115]. Rates of oxygen loss influence iron plaque formation [115], and vary between rice cultivars[116, 117]. Studies conducted over the last forty years are inconsistent on whether iron plaque prevents or enhances arsenic uptake by plants[111], and the hypothesis that arsenic influences the quality and amount of the iron plaque[113]. Profound differences in mineral composition and quantity of laboratory-created iron plaques has been demonstrated experimentally[111], which may have contributed to these inconsistencies.

5.1. Microbial activity

Microbes directly and indirectly influence arsenic speciation in rhizosphere soil, and are widely considered to play a key role in arsenic biogeochemistry[118]. Under certain nutrient-limited conditions, microbes actively weather minerals to access nutrients for cellular growth, which releases arsenic[35], as well as creating abiotic conditions that induce changes in arsenic speciation via production of organic acids, polysaccharides and ligands. Soil microorganisms can strongly affect soil redox, regulating arsenic release into pore water[119]. A number of strains of bacteria have also been shown to contribute to the formation of arsenic minerals by using arsenic as a terminal electron acceptor, such as *Desulfosporosinus auripigmentum*[120], *Desulfovibrio* strain Ben-RB[121], *Shewanella oneidensis*[122] and *S. putrefaciences* CN32[123]. These microorganisms also differ in their capabilities for liberating arsenic from specific arsenate-bearing minerals[119].

Microbial transformation can mobilize arsenic by converting inorganic to organic forms, including MMA and DMA[124, 125]. Plants translocate organic arsenicals from roots to the (frequently edible) above-ground parts more efficiently than inorganic arsenic[126-128] (See Section 6), therefore microbial transformation to organic arsenicals can increase human dietary exposure.

Plants, green algae and microbes can all enzymatically transform arsenic species[124, 129], but methylated forms of arsenic detected in plants are a product of rhizosphere bacteria; plants cannot methylate arsenic[78, 124, 130, 131]. The genomes of more than 85 arsenic-metabolizing archaea and bacteria have been sequenced for genes involved in arsenic metabolism[132]. In bacteria, archea and fungi, arsenic methylation is catalyzed by homologs of arsM, (See Section 4)[124]. Resistance to arsenite and arsenate exists in nearly all microbes, which also confers the ability to transform arsenate into volatile arsine gases[133], a particularly effective way of removing arsenic.

Profiling the transcriptome, proteome and metabolome of arsenic contaminated soils offers way of understanding microbially-mediated rhizosphere arsenic processes[132]. This approach measures the presence and expression of specific genes, rather than attempting to isolate and study the microbes that carry them, 98% of which – it is estimated - do not grow in culture[134]. Microbially mediated arsenic metabolic processes that play a major role in arsenic cycling in agronomic systems include arsenite oxidation (via the *aio* genes), arsenate respiration (via the *arr* genes), arsenate reduction (via the *ars* genes) and arsenite methylation (via the *arsM* genes)[135]. Interested readers are referred to the recent excellent work of Andres and Bertin[132] for a comprehensive review of this subject. Microbially mediated redox processes strongly influence arsenic uptake in rice, involving *aioA*, *arsC* and *arrA* [124], with pH emerging as an important factor in the distribution of microbes in paddy soils. Testing a variety of soils has shown that bacteria possessing the *arsM* gene for methylating arsenic are widespread and phylogenetically diverse, and even in paddy soils with low concentrations of arsenic, genes for arsenic metabolism are abundant[124].

6. Arsenic and crop plants

Much of our understanding about the physiological mechanisms of arsenic uptake in plants comes from the study of a limited number of species. Called *model plant* species, they are extensively studied, well described, easy to grow, and the results can be compared between studies. The understanding is that the information gained from studying model plants is applicable to other plant species. From a genetic perspective, orthologous genes exist in different plant species that have evolved from a common ancestral gene, and they usually retain the same function. Characterization of arsenic-related genes in a model plant strongly suggests that they exist and perform similar functions in other species. Caveats to this are their levels of expression, which makes some plants more adept at accumulating arsenic than others. In this section, much of the knowledge gained on arsenic uptake and metabolism of plants comes from the study of mouse-eared pennycress (also called thale cress or rockcress) (*Arabidopsis thaliana* Heynh.*)* and rice (*Oryza sativa* L.); model plants with fully sequenced genomes. These species represent dicotyledonous (e.g. flowers, vegetables, deciduous trees) and monocotyledonous plant species (e.g. grasses, palm trees) respectively, thereby representing much of the edible crop species. An exception to this is the study of the arsenic hyperaccumulating fern

(Chinese Brake fern, *Pteris vittata*), a seedless plant that is able to accumulate up to 22,630 mg/kg (dry weight) arsenic in its fronds[136].

6.1. Phytotoxicity of arsenicals

Arsenic is toxic to plants[137]. Despite lower acute human toxicity of the organic arsenicals (median lethal dose is 700-1,600 mg/kg and 700-2,600 mg/kg for MMA and DMA respectively compared to 10-20 mg/kg for inorganic forms)[138] no one form of arsenic is consistently more toxic to plants[139]. Soybean yields are affected when tissue arsenic levels exceed 1 mg/kg, and 4 mg/kg limits cotton yields[140], whereas in barley tissue concentrations of 20 mg/kg inhibited growth[141]. Higher yield-limiting arsenic levels have been recorded in rice: 20-100 mg/kg in above ground biomass, and 1000 mg/kg in root tissue[142]. By contrast, potatoes (*Solanum tuberosum* L.) suffered no growth inhibition in soils containing 290 mg/kg arsenic[71]. In some plants species, organic forms are more toxic than inorganic, for example in rice (order of toxicity: MMA > arsenite > arsenate = DMA)[143], and in smooth cordgrass (*Spartina alterniflora* Loisel) (DMA = MMA > arsenite > arsenate)[144].

Plants vary in their tolerance to arsenic, and the stress response differs for each arsenic species[145-147]. The chemical similarities between arsenate and phosphate means that arsenic can replace phosphate in biomolecules like ATP (adenosine triphosphate, a molecule used for intercellular energy transfer), with negative impacts on growth and metabolism[148]. In rice in particular, DMA and MMA induce straighthead disease (arsenic-associated straighthead disease), significantly lowering yield of certain rice varieties[66]. Straighthead is a physiological disorder of rice characterized by sterile florets, which remain upright at maturity instead of bending over under the weight of the filled grain. The exact cause of straighthead is unknown, but consistent flooding, low soil pH, high iron availability and high organic matter content have all been implicated in naturally-occurring straighthead disease[66]. Arsenic's suspected role in straighthead comes from observations of more frequent outbreaks in rice grown in soil where arsenical herbicides such as monosodium methanearsonate (MSMA) – used in cotton production in the USA – have been historically applied.

6.2. Arsenic uptake mechanisms

In magnitude, plants take up arsenicals from the soil in the order arsenite > arsenate > DMA > MMA[149, 150)], with the various arsenic species entering via different root membrane transport proteins in the root plasma membrane that allow ions and molecules to cross with varying levels of selectivity, or target specificity. Similarities in chemical structure between arsenate and phosphate, and between arsenite and silicic acid, govern their entry into root cells. Arsenate enters root cells through phosphate transporters (the Phosphate Transporter 1 family of proteins; PHT1) in both the model plant *Arabidopsis thaliana*[151, 152] and in rice[153-155] (Figure 1). In rice, Low Silicon 1 (OsLsi1) and OsLsi2 are silicic acid transporters and arsenite, MMA^{V}, and DMA^{V} are among their unintended targets[156, 157]. These Nodulin 26-like Intrinsic Proteins (NIPs)[158], which are members of the aquaporin water channel superfamily of proteins[159] embedded in the exodermal cell membranes of rice roots, move arsenic from the soil into the vascular system for distribution to the stem and leaves. OsLsi2 works in tandem with OsLsi1 to transport arsenite inward toward the xylem[160, 161] (vascular tissue that conducts water and dissolved nutrients up from the roots). The arsenic uptake specificity of OsLsi1 is arsenite >> MMA > DMA[158]. These bidirectional NIP transport proteins also efflux arsenite back in to the soil, but since OsLsi1 effluxes only 15-20% of the arsenite in roots cells[162], there may be other unidentified arsenite efflux transporters contributing to this process.

6.3. Arsenic transport and metabolism in plants

Transport of arsenite into the xylem for delivery to the shoot is less well characterized than its uptake from the soil. Arsenic is transported to the grain mainly via the phloem[126-128] (vascular tissue that conducts sugars and metabolic products from the leaves), by transporters in the nodes[163], but their characterization is still in the early stages. Transporters for myo-inositol (Inositol Transporter 2 and 4); an important sugar for developing rice grains, also transport arsenite into the phloem companion cells[164, 165]. In Arabidopsis, INT2 or AtINT4 load about 45-64% arsenite into the grain[166]. The identity of transporters that move arsenite out of the phloem and into the grain are also unknown, but manipulating the target specificity of the INT genes might show promise in molecular genetic or plant breeding mitigation efforts as a way to prevent arsenite from reaching the grain.

Despite having a lower affinity for transporters into the plant than the inorganic forms, organic arsenic species are more efficiently transported towards the shoot than inorganic forms[149, 150] because they are not complexed by phytochelatins (PCs); sulphydryl-rich glutathione (GSH) polymers[167, 168]. Likewise, in broad beans (*Vicia faba* L.) grown in a soil containing 90% inorganic arsenic, DMA and MMA were the dominant arsenic forms in the bean (68%)[169]. In root vegetables, carrot (*Daucus carota* L.) and beet (*Beta vulgaris* L.) grown on arsenic-contaminated soils, arsenic forms were predominantly inorganic, but for beets in particular were not readily identified using the typical standards (arsenate, arsenite, MMA and DMA)[170].

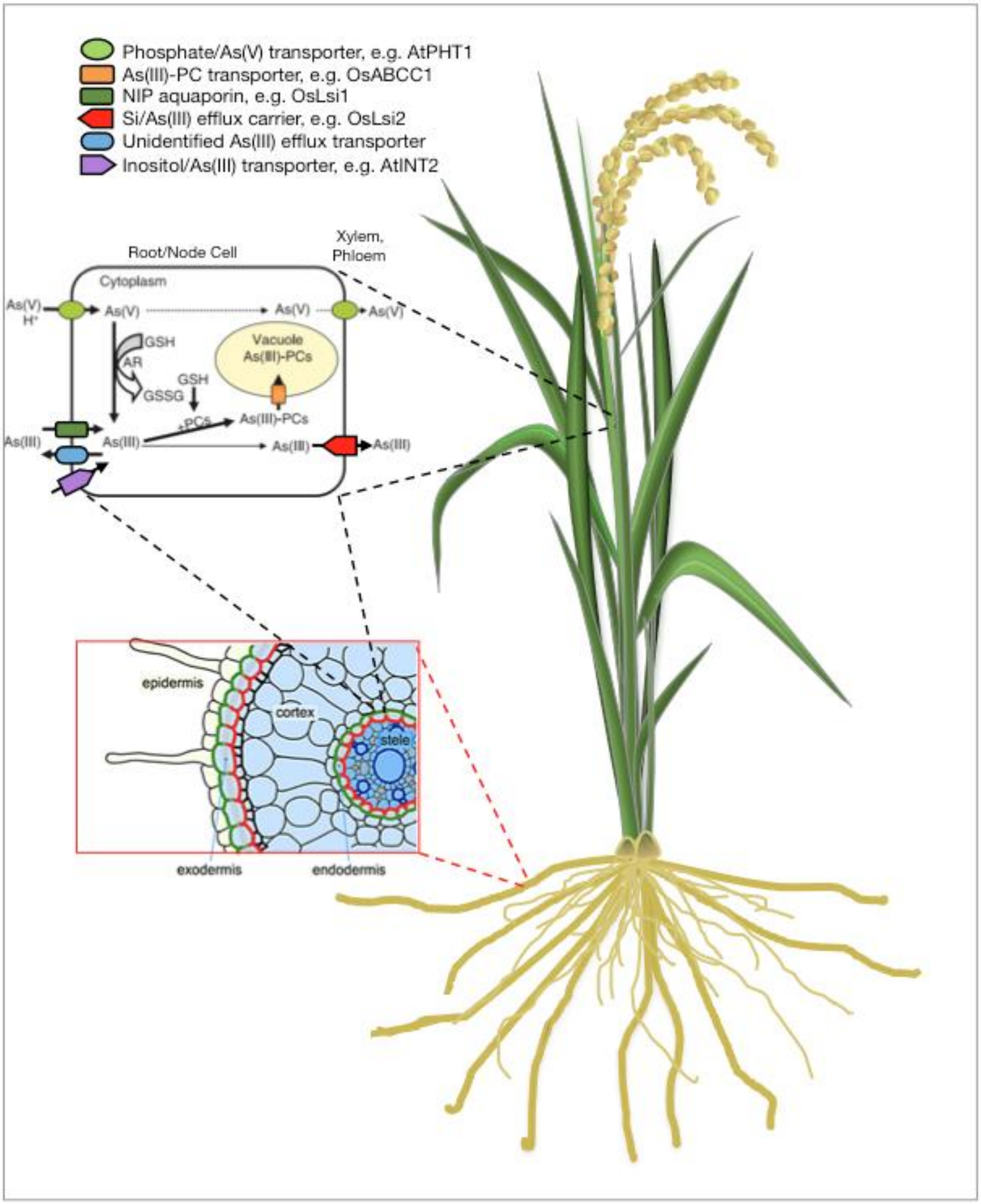


Figure 1. Generalized diagram of arsenic uptake, transport and metabolism in plants. GSH, glutathione; AR, arsenate reductase; GSSG, oxidized glutathione; PC, phytochelatin. Modified from Zhao et al.[171] and Ma et al.[172].

The arsenic species composition of rice grain is influenced by the arsenic transport rate of the particular cultivar[173, 174]. Rice cultivars currently grown in the USA have an arsenic speciation split approximately equally between inorganic arsenic and DMA, while cultivars grown in Bangladesh contain mostly inorganic arsenic[174]. While lower inorganic arsenic in rice grain seems favorable for avoiding human health effects, the assumed safety of DMA is contentious[175], being based on acute toxicity data, and not on genotoxicity or carcinogenicity, which are equally relevant in long term safety considerations.

Arsenic detoxification inside cells uses a multi-step process beginning with reduction of arsenate to arsenite using an arsenate reductase enzyme[176, 177]. In Arabidopsis, the protein High Arsenic Content1 (HAC1; also called Arsenate Reductase QTL1; ARQ1) reduces arsenate[177]. Even though arsenite is more toxic than arsenate[158, 178, 179], it is hypothesized that ancestral organisms to plants were exposed almost exclusively to arsenite before atmospheric oxygen enabled arsenate formation[180], and this mechanism persisted through natural selection. Arsenite is then complexed by PCs, and transported in to the vacuole[167] via ATP Binding Cassette tranporters[181, 182]. This process depletes glutathione availability, rendering the plant more susceptible to other oxidative stresses, which inhibits photosynthesis, pigment production, and the integrity of cell membranes[183-186].

7. Limiting arsenic uptake by crops

7.1. Water management

Although the traditional method for cultivating rice involves flooding leveled, tilled fields before or shortly after planting germinated seedlings, flooded soil is not a biological requirement of rice plants. Flooding is used for weed and vermin control, for mobilization of key nutrients such as iron, phosphate and zinc, and importantly, flooding discourages the buildup of root nematodes over multiple years of rice growth. As mentioned earlier, flooded conditions mobilize soil-bound arsenic through reductive dissolution of Fe (oxyhydr)oxides, and the reduction of arsenate to the more mobile arsenite[187]. Water management strategies that involve periods of oxic soil conditions can decrease arsenic uptake in rice by limiting dissolution of arsenic. Rice grown in non-flooded or aerobic conditions has a lower yield than intermittently or constantly flooded rice[188-190]. Intermittent flooding (flooding maintained until full tillering, followed by intermittent irrigation) is a promising management technique to reduce arsenic levels, and can potentially produce higher grain yields than either non-flooded or constantly flooded conditions[191]. However, oxic conditions increase cadmium concentrations in the grain when grown in acidic soils[191-193], and cadmium is also a highly toxic metal. The observed increases in cadmium were also a cultivar-specific trait, but the increase in cadmium uptake between rice grown under aerobic conditions were approximately an order of magnitude greater than their flooded counterparts. Pot experiments suggest that water management strategies implemented during the heading period of rice growth (when the rice panicle has emerged from the stem and is fully visible, just before flowering) can regulate both arsenic and cadmium concentration in the grain[190, 192].

7.2. Amendment and fertilization practices

Soil amendment involves incorporating substances into the plow layer that either add missing nutrients, reduce the bioavailability of existing potentially toxic substances (to prevent crop uptake), or both. Soil amendments that have shown potential in reducing arsenic uptake by plants include iron-, and silica-based additives. The use of iron-based amendments increases in the concentration of free iron oxide in the soil, retarding the release of arsenite from the solid phase into soil solution, (mentioned in Section 4.1 and discussed in Section 5), whereas silica fertilization inhibits arsenic uptake by competitive inhibition at the plant root surface while adding an essential nutrient.

Zero valent iron powder (90% iron) and iron oxide (56% iron) incorporation prevented uptake of arsenic in to the grain of rice grown on soil containing 39.5 mg/kg total arsenic by approximately 45%, and corresponded with a reduction in bioavailable arsenic in the soil[194]. Amendment with iron oxides (at a rate of 2%) was also more effective at reducing grain arsenic than phosphate amendment[195]. Amendments have also been used in combination with water management strategies to try and reduce both arsenic and cadmium concentrations in rice simultaneously[193], without success. Reduction of arsenic in the grain was achieved with iron oxide addition and constant flooding, whereas cadmium reduction was achieved with converter furnace slag addition and rain water management (no irrigation after midseason drainage until harvest).

A combination of ethylenediaminetetraceetic acid ferric sodium salt (iron EDTA) and calcium peroxide was effective for reducing arsenic uptake by vegetable crops (lettuce, Chinese cabbage and radish) from soils containing 14 mg/kg total arsenic[196], again by increasing amorphous aluminum and iron oxides. It is likely that this this level of arsenic contamination would be deemed too high for commercial vegetable production, so these amendments may only be feasible for use in private vegetable gardens. Questions remain about whether iron oxide amendment application only temporarily reduces arsenic bioavailability[197]. In addition, the suitability for arsenic immobilization is highest at the lower soil pH range, and is strongly affected by soil phosphorus concentration, which strongly competes with arsenic.

Rice plants take up high concentrations of silica, constituting up to 10% of dry matter in the straw and husk of the plant[198]. As mentioned earlier (Section 6) the silicon membrane transporter (Lsi1) is the main route of arsenite entry in to rice root cells, and provision of silicon causes competitive inhibition of arsenite uptake. Increasing silicon availability in the soil also reduces the expression of the Lsi1 transporter in the plant, which further decreases the potential for arsenic uptake. Fertilization of rice paddy soils with silicon is a potential mitigation strategy for preventing or reducing arsenic uptake by rice through competitive inhibition of arsenite uptake[199]. The use of synthetic silicon fertilizers, such as calcium silicate or silica gel is prohibitively expensive for smallholder farmers in developing countries, however reusing the silicon-rich parts of the rice plant that remain after harvesting and grain processing may provide a sustainable solution that also addresses the ongoing issue of silicon depletion of the soil[198]. Soil incorporation of fresh rice husks, or the ash that remains after burning the husk and straw for energy (which is a common practice for smallholder farmers), can provide silicon without increasing methane production and decreases either total or inorganic arsenic in rice grain[200]. Despite the potential of soil amendment with iron oxides or silica to reduce arsenic bioavailability or prevent plant uptake of arsenic, the high cost of these amendments inevitably prevents their use, especially by smallholder farmers. Large rice producers in the US or Europe have not so far adopted widespread use of these soil amendments to reduce rice grain arsenic concentrations. It is also reasonable to assume that use of expensive soil amendments would drive up the cost of rice. Their lack of use may also be attributable to the fact that iron amendments are essentially untested in a diverse range of large-scale agricultural settings and their performance will vary between soil types. In non-rice agricultural systems arsenic is tightly bound to the solid phase; significant crop uptake from oxidized soil is likely to be a result of extreme contamination, in which case effective mitigation is restricted to redirecting land use away from edible crops. In systems subject to periodic flooding, improving drainage remains the best mitigation strategy.

7.3. Mitigation using plant breeding approaches

The development of crops that accumulate high levels of arsenic and yet remain healthy, while preventing arsenic from reaching the edible grain is thought to hold great potential as a strategy for reducing human exposure to dietary arsenic. The use of molecular genetics techniques such as alterations in gene expression characteristics, gene editing to alter target specificity, or alternately, using traditional plant breeding techniques are both tangible approaches. Both use knowledge of the arsenic uptake and tolerance characteristics of plants to develop varieties with desired characteristics. These characteristics include lower arsenic uptake[201], higher arsenite efflux[202] and increased vacuolar arsenic sequestration[203]. For instance, many rice cultivars have now been screened to identify those that accumulate lower levels of arsenic in their grain and efforts are underway to identify the genes underlying this trait[201, 204]. Overexpressing Arabidopsis ABC-type transporters that sequester arsenite-PC complexes in the cell vacuole results in plants able to grow in otherwise toxic concentrations of arsenic[182]. Conversely, knocking out the function of the related rice ABC transporter OsABCC1 results in higher levels of grain arsenic. The OsABCC1 transporter limits arsenic transport to grains by sequestering arsenic in the vacuoles of the phloem companion cells directly connected to the grain. By combining what we have learned from the overexpression studies in Arabidopsis and the loss-of-function study in rice, overexpression of OsABCC1 can be used as a strategy to breed arsenic tolerance and low-arsenic accumulating rice cultivars. Another promising strategy is based on expressing the arsenate efflux transporter from yeast (*Saccharomyces cerevisiae*) in rice, which can reduce arsenic accumulation in brown rice by 20%. A less successful idea to methylate sodium arsenite to DMA by expressing an algal arsM gene in Arabidopsis resulted in lethal phytotoxicity[205], suggesting that arsenic methylation in plants can only be an effective detoxification strategy if volatile arsines are the end point of the methylation.

8. Conclusions

The discovery of arsenic in staple foods, beverages and other products has increased awareness and stimulated research on the sources and the processes involved. The information brought together here illustrates the numerous geochemical and biological processes that influence the movement of arsenic into the food supply. It is clear there must be strategies for preventing arsenic exposure, that operate in both the short term – to protect consumers from existing contamination – and in the long term, to prevent further contamination. This requires government regulation on the permissible levels of arsenic food, with lower levels for infant foods (see Nachman et al, this issue), which must work in tandem with long term goals to address arsenic in agricultural soils, actively prevent further inputs and identify contaminated areas for mitigation. Our recommendations are that the information in this review is used to inform a reconsideration and a unification of regulations on the action levels of agricultural soil arsenic, which in the USA for example, exist only at the state level, vary widely from state to state, and have no formal channels of enforcement. We recommend that educating the community and garnering their support and involvement for lowering exposure to arsenic through food is an approach already shown to hold enormous potential. Direct involvement of the commercial rice growing community in research and development of arsenic mitigation strategies and amendments is needed. Much effort has been given to short-term, greenhouse-scale testing of amendment formulations that will ultimately be too expensive, impractical, or ineffective in the long term. Community-based participatory research should extend to the agricultural community, leading to partnerships that will make longer-term field-scale testing of mitigation strategies accessible. Feasibility should be the first consideration in arsenic mitigation research. Community outreach efforts targeted to commercial growers or the home gardener specifically must raise awareness of the significance and potential impacts of former land uses, encouraging testing for the presence of arsenic in the soil and educating growers on crops shown to accumulate arsenic in their edible parts. Information gathering on former arsenic input into the soil from pesticides and from proximity to various waste sites is of paramount importance, and will allow monitoring and mitigation to be targeted to where it is needed most. Currently there is no readily available source of soil arsenic concentration information at a sufficient resolution to inform commercial producers or homeowners: this information needs to be accessible to everyone, everywhere. Going forward, management and remediation of arsenic contaminated soils is essential both for human health and food security, and innovative technologies are urgently needed that will expedite this process. Innovative solutions such as the use of rice husks to add silicon to the soil to offset arsenic uptake, and the use of cultivars with low-arsenic accumulating characteristics point the way forward for sustainable solutions.

Acknowledgements

This paper, a product of the Collaborative on Food with Arsenic and associated Risk and Regulation (C-FARR), is supported by the Dartmouth College Toxic Metals Superfund Research Program through funds from the National Institute of Environmental Health Sciences of the National Institutes of Health under Award Number 1R13ES026493-01 to C. Chen and Award Number P42ES007373 to B.

Stanton, and the Children's Environmental Health and Disease Prevention Research Center at Dartmouth through funds from the National Institute of Environmental Health Sciences of the National Institutes of Health under Award Number P01ES022832 to M. Karagas. Although EPA contributed to this article, the research presented was not performed by or funded by EPA and was not subject to EPA's quality system requirements. Consequently, the views, interpretations, and conclusions expressed in this article are solely those of the authors and do not necessarily reflect or represent EPA's views or policies. TP was supported by funds from the National Institute of General Medical Sciences Center for Biomedical Research Excellence (P20GM104416). The authors disclose that there are no actual or potential conflicts of interest.

Bibliography

1. Norton, G.J., et al., *Variation in grain arsenic assessed in a diverse panel of rice (Oryza sativa) grown in multiple sites.* New Phytologist, 2012. **193**(3): p. 650-664.
2. Carlin, D.J., et al., *Arsenic and Environmental Health: State of the Science and Future Research Opportunities.* Environ Health Perspect, 2015.
3. Bodwell, J.E., L.A. Kingsley, and J.W. Hamilton, *Arsenic at very low concentrations alters glucocorticoid receptor (GR)-mediated gene activation but not GR mediated gene repression; complex dose response effects are closely correlated with levels of activated GR and require a functional GR DNA binding domain.* Chemical Research in Toxicology, 2004. **17**: p. 1064-1076.
4. Davis, M.A., et al., *Rice consumption and urinary arsenic concentrations in US children.* Environmental Health Perpectives, 2012. **120**: p. 1418–1424.
5. WHO. *Arsenic*. 2016 [cited 2016 November 22, 2016]; International Programme on Chemical Safety. Health Impacts of Chemicals:]. Available from: http://www.who.int/ipcs/assessment/public_health/arsenic/en/.
6. Union, E. *Commission Regulation (EU) 2015/1006 of 25 June 2015 amending Regulation No. 1881/2006 as regards maximum levels of inorganic arsenic in foodstuffs*. 2015 [cited 2016 November 22, 2016]; Free access to EU law (treaties, directives, regulations, decisions, consolidated legislation), preparatory acts, case-law, international agreements, EFTA focuments, summaries of EU legislation and other documents.]. Available from: http://eur-lex.europa.eu/eli/reg/2015/1006/oj.
7. Moon, K., E. Guallar, and A. Navas-Acien, *Arsenic exposure and cardiovascular disease: an updated systematic review.* Curr Atheroscler Rep, 2012. **14**(6): p. 542-55.
8. Caldwell, K.E., et al., *Prenatal arsenic exposure alters the programming of the glucocorticoid signaling system during embryonic development.* Neurotoxicol Teratol, 2014. **47C**: p. 66-79.
9. Yamamura, S., et al., *Drinking water guidelines and standards. In United Nations Synthesis Report on Arsenic in Drinking water.* . 2001, United Nations: United Nation, Geneva. p. 18.

10. FDA, U., *FDA proposes limit for inorganic arsenic in infant rice cereal*, L. Sucher, Editor. 2016, US Food and Drug Administration.

11. Japan, M.o.E.G.o. *Environmental Quality Standards for Soil Pollution*. Water/Soil/Ground Environment 2016 [cited 2016 12 December].

12. Henke, K., *Appendix E: Regulation of Arsenic: A Brief Survey and Bibliography*, in *Arsenic*, K. Henke, Editor. 2009, John Wiley & Sons, Ltd. p. 545-557.

13. Adomako, E.E., et al., *Enhanced transfer of arsenic to grain for Bangladesh grown rice compared to US and EU.* Environment International, 2009. **35**(3): p. 476-479.

14. Lu, Y., et al., *Baseline Soil Variation Is a Major Factor in Arsenic Accumulation in Bengal Delta Paddy Rice.* Environmental Science & Technology, 2009. **43**(6): p. 1724-1729.

15. Williams, P.N., et al., *Greatly enhanced arsenic shoot assimilation in rice leads to elevated grain levels compared to wheat and barley.* Environmental Science & Technology, 2007. **41**(19): p. 6854-6859.

16. Norton, G., et al., *Arsenic Speciation and Localization in Horticultural Produce Grown in a Historically Impacted Mining Region.* Environmental Science & Technology, 2013. **47**(12): p. 6164-6172.

17. Koljonen, T., et al., *12th International Geochemical Exploration Symposium and the 4th Symposium on Methods of Geochemical Prospecting Geochemical Atlas of Finland: preliminary aspects.* Journal of Geochemical Exploration, 1989. **32**(1): p. 231-242.

18. Geiszinger, A., W. Goessler, and W. Kosmus, *Organoarsenic compounds in plants and soil on top of an ore vein.* Applied Organometallic Chemistry, 2002. **16**(5): p. 245-249.

19. IMA, *The new International Mineralogical Association list of minerals. A work in progress.* . 2014, International Mineralogical Society: http://www.ima-mineralogy.org/Minlist.htm.

20. Lado, L.R., T. Hengl, and H.I. Reuter, *Heavy metals in European soils: A geostatistical analysis of the FOREGS Geochemical database.* Geoderma, 2008. **148**(2): p. 189-199.

21. Shacklette, H.T. and J.G. Boerngen, *Element Concentrations in soils and other surficial materials of the Conterminous United States*. 1984, US Geological Survey: US Government Printing Office, Washington, DC. p. 105.

22. Smith, D.B., et al., *Geochemical and mineralogical maps for soils of the conterminous United States: U.S.*, U.G. Survey, Editor. 2014. p. 386.

23. USGS. *Mineral Resources On-Line Spatial Data: National Geochemical Survey database*. [Website] 2016 September 3 2014 [cited 2016 July 18]; National scale geochemical analysis of stream sediments and soils in the US, from existing data, reanalysis of existing samples and new sampling.]. Available from: http://mrdata.usgs.gov/geochemistry/ngs.html.

24. Young, M. and A. Donald, *A guide to the Tellus data.* 2013, Belfast, UK: Geological Survey of Northern Ireland. 233.

25. Smedley, P.L. and D.G. Kinniburgh, *A review of the source, behaviour and distribution of arsenic in natural waters.* Applied Geochemistry, 2002. **17**(5): p. 517-568.

26. Saunders, J.A., et al., *Natural arsenic contamination of Holocene alluvial aquifers by linked tectonic, weathering, and microbial processes.* Geochemistry Geophysics Geosystems, 2005. **6**.

27. Mestrot, A., et al., *Field Fluxes and Speciation of Arsines Emanating from Soils.* Environmental Science & Technology, 2011. **45**(5): p. 1798-1804.

28. Lawgali, Y.F. and A.A. Meharg, *Levels of Arsenic and Other Trace Elements in Southern Libyan Agricultural Irrigated Soil and Non-irrigated Soil Projects.* Water Quality Exposure and Health, 2011. **3**(2): p. 79-90.

29. Melegy, A., et al., *Weathering fluxes of arsenic from a small catchment in Slovak Republic.* Environmental Earth Sciences, 2011. **64**(2): p. 549-555.

30. Drahota, P., et al., *Weathering and erosion fluxes of arsenic in watershed mass budgets.* Science of the Total Environment, 2006. **372**(1): p. 306-316.

31. Huang, J.H. and E. Matzner, *Biogeochemistry of organic and inorganic arsenic species in a forested catchment in Germany.* Environmental Science & Technology, 2007. **41**(5): p. 1564-1569.

32. CEH. *United Kingdom Pollutant Deposition Maps*. 2008 2016]; Database]. Available from: http://www.pollutantdeposition.ceh.ac.uk/content/heavy-metals.

33. UKSO. *NSI topsoil Arsenic map*. 2016 [cited 2016 August 11]; Topsoil arsenic map of England and Wales]. Available from: http://www.ukso.org/nsi/Arsenic.html.

34. Mikutta, C. and J.J. Rothwell, *Peat Bogs as Hotspots for Organoarsenical Formation and Persistence.* Environmental Science & Technology, 2016. **50**(8): p. 4314-4323.

35. Mailloux, B.J., et al., *Microbial Mineral Weathering for Nutrient Acquisition Releases Arsenic.* Applied and Environmental Microbiology, 2009. **75**(8): p. 2558-2565.

36. Williams, P.N., et al., *Organic Matter-Solid Phase Interactions Are Critical for Predicting Arsenic Release and Plant Uptake in Bangladesh Paddy Soils.* Environmental Science & Technology, 2011. **45**(14): p. 6080-6087.

37. Murphy, E.A. and M. Aucott, *An assessment of the amounts of arsenical pesticides used historically in a geographical area.* The Science of The Total Environment, 1998. **218**(2-3): p. 89-101.

38. Lasky, T., et al., *Mean total arsenic concentrations in chicken 1989-2000 and estimated exposures for consumers of chicken.* Environmental Health Perspectives, 2004. **112**(1): p. 18-21.

39. Nachman, K.E., et al., *Roxarsone, Inorganic Arsenic, and Other Arsenic Species in Chicken: A US-Based Market Basket Sample.* Environmental Health Perspectives, 2013. **121**(7): p. 818-824.

40. Fendorf, S. and B.D. Kocar, *Chapter 3 Biogeochemical Processes Controlling the Fate and Transport of Arsenic: Implications for South and Southeast Asia*, in *Advances in Agronomy*. 2009, Academic Press. p. 137-164.

41. Bowell, R.J., et al., *The Environmental Geochemistry of Arsenic - An Overview -.* Arsenic: Environmental Geochemistry, Mineralogy, and Microbiology, 2014. **79**: p. 1-16.

42. Craw, D. and R.J. Bowell, *The Characterization of Arsenic in Mine Waste*, in *Arsenic: Environmental Geochemistry, Mineralogy, and Microbiology*, R.J. Bowell, et al., Editors. 2014. p. 473-505.

43. Bowell, R.J., et al., Reviews in mineralogy and geochemistry, 2014. **79**: p. 635.

44. Bowell, R.J., et al., *The Environmental Geochemistry of Arsenic - An Overview*, in *Arsenic: Environmental Geochemistry, Mineralogy, and Microbiology*, R.J. Bowell, et al., Editors. 2014. p. 1-16.

45. Ramirez-Andreotta, M.D., et al., *Home gardening near a mining site in an arsenic-endemic region of Arizona: Assessing arsenic exposure dose and risk*

via ingestion of home garden vegetables, soils, and water. Science of the Total Environment, 2013. **454**: p. 373-382.

46. Ramirez-Andreotta, M.D., et al., *A greenhouse and field-based study to determine the accumulation of arsenic in common homegrown vegetables grown in mining-affected soils.* Science of the Total Environment, 2013. **443**: p. 299-306.

47. Bertoldi, D., et al., *Arsenic present in the soil-vine-wine chain in vineyards situated in an old mining area in Trentino, Italy.* Environmental Toxicology and Chemistry, 2013. **32**(4): p. 773-779.

48. Ma, L., et al., *Arsenic speciation in locally grown rice grains from Hunan Province, China: Spatial distribution and potential health risk.* Science of The Total Environment, 2016. **557–558**: p. 438-444.

49. Williams, P.N., et al., *Occurrence and Partitioning of Cadmium, Arsenic and Lead in Mine Impacted Paddy Rice: Hunan, China.* Environmental Science & Technology, 2009. **43**(3): p. 637-642.

50. Zhu, Y.G., et al., *High percentage inorganic arsenic content of mining impacted and nonimpacted Chinese rice.* Environmental Science & Technology, 2008. **42**(13): p. 5008-5013.

51. Kolker, A., et al., *Arsenic in coal*, in *Fact Sheet*. 2006. p. 4.

52. EPA. *2016*. Coal Ash Basics [Web Page] EPA June 7, 2016; Available from: https://www.epa.gov/coalash/coal-ash-basics.

53. Feuerborn, H.-J. *Coal Combustion Products in Europe - an update on Production and Ultilisation Standardisation and Regulation.* in *World of Coal Ash (WOCA) Conference*. 2011. Denver, CO, USA.

54. Yager, J.W., T. Greene, and R.A. Schoof, *Arsenic relative bioavailability from diet and airborne exposures: Implications for risk assessment.* Science of the Total Environment, 2015. **536**: p. 368-381.

55. Schmidt, C., *COAL ASH SPILL STILL A PROBLEM.* Chemical & Engineering News Archive, 2010. **88**(49): p. 13.

56. Schumann, A.W. and M.E. Sumner, *Plant nutrient availability from mixtures of fly ashes and biosolids.* Journal of Environmental Quality, 1999. **28**(5): p. 1651-1657.

57. Jackson, B.P., et al., *Trace element solubility from land application of fly ash/organic waste mixtures.* Journal of Environmental Quality, 1999. **28**(2): p. 639-647.

58. Schumann, A.W. and M.E. Sumner, *Formulation of environmentally soundwaste mixtures for land application.* Water Air and Soil Pollution, 2004. **152**(1-4): p. 195-217.

59. Yokel, J. and D.A. Delistraty, *Arsenic, lead, and other trace elements in soils contaminated with pesticide residues at the Hanford site (USA).* Environmental Toxicology, 2003. **18**(2): p. 104-114.

60. Embrick, L.L., et al., *Characterization of lead and arsenic contamination at Barber Orchard, Haywood County, NC.* Microchemical Journal, 2005. **81**(1): p. 117-121.

61. Renshaw, C.E., et al., *Impact of land disturbance on the fate of arsenical pesticides.* Journal of Environmental Quality, 2006. **35**(1): p. 61-67.

62. Peryea, F.J. and T.L. Creger, *VERTICAL-DISTRIBUTION OF LEAD AND ARSENIC IN SOILS CONTAMINATED WITH LEAD ARSENATE PESTICIDE-RESIDUES.* Water Air and Soil Pollution, 1994. **78**(3-4): p. 297-306.

63. Robinson, G.R., et al., *Assessment of contamination from arsenical pesticide use on orchards in the Great Valley region, Virginia and West Virginia, USA.* Journal of Environmental Quality, 2007. **36**(3): p. 654-663.

64. Peryea, F.J. and R. Kammereck, *Phosphate-enhanced movement of arsenic out of lead arsenate-contaminated topsoil and through uncontaminated subsoil.* Water Air and Soil Pollution, 1997. **93**(1-4): p. 243-254.

65. Bednar, A.J., et al., *Presence of organoarsenicals used in cotton production in agricultural water and soil of the southern United States.* Journal of Agricultural and Food Chemistry, 2002. **50**(25): p. 7340-7344.

66. Rahman, M.A., et al., *Straighthead disease of rice (Oryza sativa L.) induced by arsenic toxicity.* Environmental and Experimental Botany, 2008. **62**(1): p. 54-59.

67. Hudak, P.F., *Distribution and sources of arsenic in the southern High Plains Aquifer, Texas, USA.* Journal of Environmental Science and Health Part a-Toxic/Hazardous Substances & Environmental Engineering, 2000. **35**(6): p. 899-913.

68. Reedy, R.C., et al., *Unsaturated zone arsenic distribution and implications for groundwater contamination.* Environmental Science & Technology, 2007. **41**(20): p. 6914-6919.

69. Ayotte, J.D., et al., *Modeling the probability of arsenic in groundwater in New England as a tool for exposure assessment.* Environmental Science & Technology, 2006. **40**(11): p. 3578-3585.

70. Feng, M., et al., *Arsenic transport and transformation associated with MSMA application on a golf course green.* Journal of Agricultural and Food Chemistry, 2005. **53**(9): p. 3556-3562.

71. Codling, E.E., R.L. Chaney, and C.E. Green, *Accumulation of Lead and Arsenic by Potato Grown on Lead-Arsenate-Contaminated Orchard Soils.* Communications in Soil Science and Plant Analysis, 2016. **47**(6): p. 799-807.

72. Codling, E.E., R.L. Chaney, and C.E. Green, *Accumulation of lead and arsenic by carrots grown on lead-arsenate contaminated orchard soils.* Journal of Plant Nutrition, 2015. **38**(4): p. 509-525.

73. McBride, M.B., et al., *Arsenic and Lead Uptake by Vegetable Crops Grown on an Old Orchard Site Amended with Compost.* Water Air and Soil Pollution, 2015. **226**(8).

74. Lim, M.P. and M.B. McBride, *Arsenic and lead uptake by Brassicas grown on an old orchard site.* Journal of Hazardous Materials, 2015. **299**: p. 656-663.

75. Zavala, Y.J. and J.M. Duxbury, *Arsenic in rice: I. Estimating normal levels of total arsenic in rice grain.* Environmental Science & Technology, 2008. **42**(10): p. 3856-60.

76. Zavala, Y.J., et al., *Arsenic in rice: II. Arsenic speciation in USA grain and implications for human health.* Environmental Science & Technology, 2008. **42**(10): p. 3861-6.

77. Williams, P.N., et al., *Variation in arsenic speciation and concentration in paddy rice related to dietary exposure.* Environmental Science and Technology, 2005. **39**(15): p. 5531-5540.

78. Lomax, C., et al., *Methylated arsenic species in plants originate from soil microorganisms.* New Phytol, 2012. **193**(3): p. 665-72.

79. Pouschat, P. and G.J. Zagury, *In vitro gastrointestinal bioavailability of arsenic in soils collected near CCA-treated utility poles.* Environmental Science & Technology, 2006. **40**(13): p. 4317-4323.

80. Townsend, T.G., et al., *Chromium, copper and arsenic concentrations in soil underneath CCA-treated wood structures. .* Soil & Sediment Contamination, 2003. **12**: p. 779-798.

81. Juhasz, A.L., J. Weber, and E. Smith, *Predicting Arsenic Relative Bioavailability in Contaminated Soils Using Meta Analysis and Relative Bioavailability-Bioaccessibility Regression Models.* Environmental Science & Technology, 2011. **45**(24): p. 10676-10683.

82. Carbonell-Barrachina, A.A., et al., *Arsenic chemistry in municipal sewage sludge as affected by redox potential and pH.* Water Research, 2000. **34**(1): p. 216-224.

83. USFDA. *Arsenic-based Animal Drugs and Poultry*. Available from: http://www.fda.gov/AnimalVeterinary/SafetyHealth/ProductSafetyInformation/ucm257540.htm.

84. Taylor, D.A., *Funky chicken - Consumers exposed to arsenic in poultry.* Environmental Health Perspectives, 2004. **112**(1): p. A50-A50.

85. Bednar, A.J., et al., *Photodegradation of roxarsone in poultry litter leachates.* The Science of The Total Environment, 2003. **302**(1-3): p. 237-245.

86. Stolz, J.F., et al., *Biotransformation of 3-nitro-4-hydroxybenzene arsonic acid (roxarsone) and release of inorganic arsenic by Clostridium species.* Environmental Science & Technology, 2007. **41**(3): p. 818-823.

87. Garbarino, J.R., et al., *Environmental fate of roxarsone in poultry litter. I. Degradation of roxarsone during composting.* Environmental Science & Technology, 2003. **37**(8): p. 1509-1514.

88. Jackson, B.P., et al., *Trace element speciation in poultry litter.* J Environ Qual, 2003. **32**(2): p. 535-40.

89. Jackson, B.P., J.C. Seaman, and P.M. Bertsch, *Fate of arsenic compounds in poultry litter upon land application.* Chemosphere, 2006. **65**(11): p. 2028-2034.

90. Rutherford, D.W., et al., *Environmental fate of roxarsone in poultry litter. part II. Mobility of arsenic in soils amended with poultry litter.* Environmental Science & Technology, 2003. **37**(8): p. 1515-1520.

91. Han, F.X., et al., *Arsenic solubility and distribution in poultry waste and long-term amended soil.* Science of the Total Environment, 2004. **320**(1): p. 51-61.

92. Fisher, D.J., L.T. Yonkos, and K.W. Staver, *Environmental Concerns of Roxarsone in Broiler Poultry Feed and Litter in Maryland, USA.* Environmental Science & Technology, 2015. **49**(4): p. 1999-2012.

93. Shah, S.B., et al., *Leaching of Nutrients and Trace Elements from Stockpiled Turkey Litter into Soil.* Journal of Environmental Quality, 2009. **38**(3): p. 1053-1065.

94. Castlehouse, H., et al., *Biotransformation and accumulation of arsenic in soil amended with seaweed.* Environ Sci Technol, 2003. **37**(5): p. 951-7.

95. Antaya, N.T., et al., *Incremental amounts of Ascophyllum nodosum meal do not improve animal performance but do increase milk iodine output in early lactation dairy cows fed high-forage diets1.* Journal of Dairy Science, 2015. **98**(3): p. 1991-2004.

96. Bowell, R.J., *Sorption of Arsenic by Iron-Oxides and Oxyhydroxides in Soils.* Applied Geochemistry, 1994. **9**(3): p. 279-286.

97. Luxton, T.P., C.J. Tadanier, and M.J. Eick, *Mobilization of arsenite by competitive interaction with silicic acid.* Soil Science Society of America Journal, 2006. **70**(1): p. 204-214.

98. Meharg, A.A. and F.J. Zhao, *Arsenic & rice*. 2012, Dordrecht: Springer Verlag. xi, 171 p.

99. Wang, P., et al., *A review on completing arsenic biogeochemical cycle: Microbial volatilization of arsines in environment.* Journal of Environmental Sciences, 2014. **26**(2): p. 371-381.

100. Turpeinen, R., M. Pantsar-Kallio, and T. Kairesalo, *Role of microbes in controlling the speciation of arsenic and production of arsines in contaminated soils.* Science of The Total Environment, 2002. **285**(1–3): p. 133-145.

101. Pongratz, R., *Arsenic speciation in environmental samples of contaminated soil.* Science of the Total Environment, 1998. **224**(1-3): p. 133-141.

102. Frohne, T., et al., *Controlled variation of redox conditions in a floodplain soil: Impact on metal mobilization and biomethylation of arsenic and antimony.* Geoderma, 2011. **160**(3-4): p. 414-424.

103. Jia, Y., et al., *Microbial Arsenic Methylation in Soil and Rice Rhizosphere.* Environmental Science & Technology, 2013. **47**(7): p. 3141-3148.

104. Mestrot, A., et al., *Arsenic volatilization in model anaerobic biogas digesters.* Applied Geochemistry, 2013. **33**: p. 294-297.

105. Edvantoro, B.B., et al., *Microbial formation of volatile arsenic in cattle dip site soils contaminated with arsenic and DDT.* Applied Soil Ecology, 2004. **25**(3): p. 207-217.

106. Liu, S., et al., *Arsenic removal from contaminated soil via biovolatilization by genetically engineered bacteria under laboratory conditions.* Journal of Environmental Sciences, 2011. **23**(9): p. 1544-1550.

107. Yang, J.K., et al., *Adsorption, oxidation, and bioaccessibility of As(III) in soils.* Environmental Science & Technology, 2005. **39**(18): p. 7102-7110.

108. Newman, E.I., *The rhizosphere: carbon sources and microbial populations.* , in *Ecological Interactions in Soil: Plants, Microbes and Animals*, A.H. Fitter, D. Atkinson, and D.J. Read, Editors. 1985, Blackwell Scientific: Oxford, UK. p. 107-121.

109. Acosta, J.A., J.M. Arocena, and A. Faz, *Speciation of arsenic in bulk and rhizosphere soils from artisanal cooperative mines in Bolivia.* Chemosphere, 2015. **138**: p. 1014-1020.

110. Wu, C., et al., *The effect of silicon on iron plaque formation and arsenic accumulation in rice genotypes with different radial oxygen loss (ROL).* Environmental Pollution, 2016. **212**: p. 27-33.

111. Seyfferth, A.L., *Abiotic effects of dissolved oxyanions on iron plaque quantity and mineral composition in a simulated rhizosphere.* Plant and Soil, 2015. **397**(1-2): p. 43-61.

112. Yamaguchi, N., et al., *Arsenic Distribution and Speciation near Rice Roots Influenced by Iron Plaques and Redox Conditions of the Soil Matrix.* Environmental Science & Technology, 2014. **48**(3): p. 1549-1556.

113. Lee, C.-H., et al., *Iron plaque formation and its effect on arsenic uptake by different genotypes of paddy rice.* Plant and Soil, 2013. **363**(1-2): p. 231-241.

114. Hossain, M.B., et al., *The effects of iron plaque and phosphorus on yield and arsenic accumulation in rice.* Plant Soil, 2009. **317**(1-2): p. 167-176.

115. Wu, C., et al., *Do radial oxygen loss and external aeration affect iron plaque formation and arsenic accumulation and speciation in rice?* Journal of Experimental Botany, 2012. **63**(8): p. 2961-2970.

116. Li, R.Y., et al., *Uptake and Accumulation Characteristics of Arsenic and Iron Plaque in Rice at Different Growth Stages.* Communications in Soil Science and Plant Analysis, 2015. **46**(19): p. 2509-2522.
117. Mei, X.Q., et al., *The effects of radial oxygen loss on arsenic tolerance and uptake in rice and on its rhizosphere.* Environmental Pollution, 2012. **165**: p. 109-117.
118. Gadd, G.M., *Metals, minerals and microbes: geomicrobiology and bioremediation.* Microbiology-Sgm, 2010. **156**: p. 609-643.
119. Wang, Y., et al., *Release and transformation of arsenic from As-bearing iron minerals by Fe-reducing bacteria.* Chemical Engineering Journal, 2016. **295**: p. 29-38.
120. Newman, D.K., et al., *Dissimilatory arsenate and sulfate reduction in Desulfotomaculum auripigmentum sp. nov.* Archives of Microbiology, 1997. **168**(5): p. 380-388.
121. Macy, J.M., et al., *Two new arsenate/sulfate-reducing bacteria: mechanisms of arsenate reduction.* Archives of Microbiology, 2000. **173**(1): p. 49-57.
122. Wang, J., et al., *Biotransformation and biomethylation of arsenic by Shewanella oneidensis MR-1.* Chemosphere, 2016. **145**: p. 329-335.
123. Smeaton, C.M., et al., *Simultaneous Release of Fe and As during the Reductive Dissolution of Pb-As Jarosite by Shewanella putrefaciens CN32.* Environmental Science & Technology, 2012. **46**(23): p. 12823-12831.
124. Jia, Y., et al., *Microbial Arsenic Methylation in Soil and Rice Rhizosphere.* Environmental Science & Technology, 2013. **47**(7): p. 3141-3148.
125. Xu, L., et al., *Speciation change and redistribution of arsenic in soil under anaerobic microbial activities.* Journal of Hazardous Materials, 2016. **301**: p. 538-546.
126. Carey, A.M., et al., *Phloem transport of arsenic species from flag leaf to grain during grain filling.* New Phytologist, 2011. **192**(1): p. 87-98.
127. Carey, A.-M., et al., *A review of recent developments in the speciation and location of arsenic and selenium in rice grain.* Analytical and Bioanalytical Chemistry, 2011. **402**: p. 3275-3286.
128. Carey, A.M., et al., *Grain unloading of arsenic species in rice.* Plant Physiology, 2010. **152**(1): p. 309-319.

129. Baker, M.D., et al., *Methylation of Arsenic by Fresh-Water Green-Algae.* Canadian Journal of Fisheries and Aquatic Sciences, 1983. **40**(8): p. 1254-1257.

130. Hansen, H.R., et al., *Identification of tetramethylarsonium in rice grains with elevated arsenic content.* Journal of Environmental Monitoring, 2011. **13**(1): p. 32-34.

131. Arao, T., et al., *Effects of Arsenic Compound Amendment on Arsenic Speciation in Rice Grain.* Environmental Science & Technology, 2011. **45**(4): p. 1291-1297.

132. Andres, J. and P.N. Bertin, *The microbial genomics of arsenic.* Fems Microbiology Reviews, 2016. **40**(2): p. 299-322.

133. Zhang, S., et al., *Cometabolic biotransformation of fenpropathrin by Clostridium species strain ZP3.* Biodegradation, 2011. **22**(5): p. 869-75.

134. Stewart, E.J., *Growing Unculturable Bacteria.* Journal of Bacteriology, 2012. **194**(16): p. 4151-4160.

135. Xiao, K.-Q., et al., *Metagenomic analysis revealed highly diverse microbial arsenic metabolism genes in paddy soils with low-arsenic contents.* Environmental Pollution, 2016. **211**: p. 1-8.

136. Ma, L.Q., et al., *A fern that hyperaccumulates arsenic (vol 409, pg 579, 2001).* Nature, 2001. **411**(6836): p. 438-U3.

137. Sharma, I., *Arsenic induced oxidative stress in plants.* Biologia, 2012. **67**(3): p. 447-453.

138. Le, X.C., et al., *Determination of monomethylarsonous acid, a key arsenic methylation intermediate, in human urine.* Environmental Health Perspectives, 2000. **108**(11): p. 1015-1018.

139. Finnegan, P.M. and W. Chen, *Arsenic toxicity: the effects on plant metabolism.* Front Physiol, 2012. **3**: p. 182.

140. Deuel, L.E. and A.R. Swoboda, *Arsenic toxicity to cotton and soybeans.* Journal of Environmental Quality, 1972. **1**(3): p. 317-320.

141. Davis, R.D., P.H.T. Beckett, and E. Wollan, *Critical levels of twenty potentially toxic elements in young spring barley.* Plant Soil, 1978. **49**: p. 395-408.

142. Adriano, D.C., *Trace Elements in Terrestrial Environments*. 2nd ed. 2001, New York: Springer Verlag. 867.

143. Marin, A.R., P.H. Masscheleyn, and W.H. Patrick, *The influence of chemical form and concentration of arsenic on rice growth and tissue arsenic concentration.* Plant and Soil, 1992. **139**: p. 175-183.

144. Carbonell-Barrachina, A.A., et al., *The influence of arsenic chemical form and concentration on Spartina patens and Spartina alterniflora growth and tissue arsenic concentration.* Plant and Soil, 1998. **198**: p. 33-43.

145. Choudhury, B., S. Chowdhury, and A.K. Biswas, *Regulation of growth and metabolism in rice (Oryza sativa L.) by arsenic and its possible reversal by phosphate.* J. Plant Interact., 2011. **6**(1): p. 15-24.

146. Shri, M., et al., *Effect of arsenic on growth, oxidative stress, and antioxidant system in rice seedlings.* Ecotoxicol. Environ. Safety, 2009. **72**(4): p. 1102-1110.

147. Chakrabarty, D., et al., *Comparative transcriptome analysis of arsenate and arsenite stresses in rice seedlings.* Chemosphere, 2009. **74**(5): p. 688-702.

148. Ullrich-Eberius, C.I., A. Sanz, and A.J. Novacky, *Evaluation of arsenate-associated and vanadate-associated changes of electrical membrane potential and phosphate transport in Lemna gibba G1.* J. Exp. Bot., 1989. **40**(210): p. 119-128.

149. Raab, A., et al., *Uptake and translocation of inorganic and methylated arsenic species by plants.* Environmental Chemistry, 2007. **4**(3): p. 197-203.

150. Finnegan, P.M. and W.H. Chen, *Arsenic toxicity: the effects on plant metabolism.* Frontiers in Physiology, 2012. **3**.

151. Shin, H., et al., *Phosphate transport in Arabidopsis: Pht1;1 and Pht1;4 play a major role in phosphate acquisition from both low- and high-phosphate environments.* Plant J, 2004. **39**(4): p. 629-42.

152. Remy, E., et al., *The Pht1;9 and Pht1;8 transporters mediate inorganic phosphate acquisition by the Arabidopsis thaliana root during phosphorus starvation.* New Phytol, 2012. **195**(2): p. 356-71.

153. Jia, H., et al., *The phosphate transporter gene OsPht1;8 is involved in phosphate homeostasis in rice.* Plant Physiol, 2011. **156**(3): p. 1164-75.

154. Ye, Y., et al., *The Phosphate Transporter Gene OsPht1;4 Is Involved in Phosphate Homeostasis in Rice.* PLoS One, 2015. **10**(5): p. e0126186.

155. Sun, S., et al., *A constitutive expressed phosphate transporter, OsPht1;1, modulates phosphate uptake and translocation in phosphate-replete rice.* Plant Physiol, 2012. **159**(4): p. 1571-81.

156. Li, R.Y., et al., *The rice aquaporin Lsi1 mediates uptake of methylated arsenic species.* Plant Physiol, 2009. **150**(4): p. 2071-80.

157. Ma, J.F., et al., *Transporters of arsenite in rice and their role in arsenic accumulation in rice grain.* Proc Natl Acad Sci U S A, 2008. **105**(29): p. 9931-5.

158. Abedin, M.J., J. Feldmann, and A.A. Meharg, *Uptake kinetics of arsenic species in rice plants.* Plant Physiol, 2002. **128**(3): p. 1120-8.

159. Pommerrenig, B., T.A. Diehn, and G.P. Bienert, *Metalloido-porins: Essentiality of Nodulin 26-like intrinsic proteins in metalloid transport.* Plant Sci, 2015. **238**: p. 212-27.

160. Mitani, N., et al., *Identification and Characterization of Maize and Barley Lsi2-Like Silicon Efflux Transporters Reveals a Distinct Silicon Uptake System from That in Rice.* Plant Cell, 2009. **21**(7): p. 2133-2142.

161. Ma, J.F., et al., *An efflux transporter of silicon in rice.* Nature, 2007. **448**(7150): p. 209-12.

162. Zhao, F.J., et al., *The role of the rice aquaporin Lsi1 in arsenite efflux from roots.* New Phytol, 2010. **186**(2): p. 392-9.

163. Song, W.Y., et al., *A rice ABC transporter, OsABCC1, reduces arsenic accumulation in the grain.* Proc Natl Acad Sci U S A, 2014. **111**(44): p. 15699-704.

164. Schneider, S., et al., *Arabidopsis INOSITOL TRANSPORTER4 Mediates High-Affinity H(+) Symport of Myoinositol across the Plasma Membrane.* Plant Physiol, 2006. **141**(2): p. 565-77.

165. Schneider, S., et al., *Arabidopsis INOSITOL TRANSPORTER2 Mediates H+ Symport of Different Inositol Epimers and Derivatives across the Plasma Membrane.* Plant Physiology, 2007. **145**(4): p. 1395-1407.

166. Duan, G.-L., et al., *Inositol transporters AtINT2 and AtINT4 regulate arsenic accumulation in Arabidopsis seeds.* Nature Plants, 2015. **2**: p. 15202.

167. Zenk, M.H., *Heavy metal detoxification in higher plants--a review.* Gene, 1996. **179**(1): p. 21-30.

168. Schmoger, M.E., M. Oven, and E. Grill, *Detoxification of arsenic by phytochelatins in plants.* Plant Physiol, 2000. **122**(3): p. 793-801.

169. Sadee, B.A., M.E. Foulkes, and S.J. Hill, *A study of arsenic speciation in soil, irrigation water and plant tissue: A case study of the broad bean plant, Vicia faba.* Food Chemistry, 2016. **210**: p. 362-370.

170. Pizarro, I., et al., *Bioaccessibility and arsenic speciation in carrots, beets and quinoa from a contaminated area of Chile.* Science of The Total Environment, 2016. **565**: p. 557-563.

171. Zhao, F.J., et al., *Arsenic uptake and metabolism in plants.* New Phytologist, 2009. **181**: p. 777-794.

172. Ma, J.F., et al., *An efflux transporter of silicon in rice.* Nature, 2007. **448**: p. 209-12.

173. Seyfferth, A., et al., *Defining the distribution of arsenic species and plant nutrients in rice (Oryza sativa L.) from the root to the grain.* Geochim. Cosmochim. Acta, 2011. **75**: p. 6655-6671.

174. Williams, P.N., et al., *Variation in arsenic speciation and concentration in paddy rice related to dietary exposure.* Environ. Sci. Technol., 2005. **39**(15): p. 5531-5540.

175. Hughes, M.F., *Arsenic toxicity and potential mechanisms of action* Toxicology Letters, 2002. **133**(1): p. 1-16.

176. Sanchez-Bermejo, E., et al., *Natural variation in arsenate tolerance identifies an arsenate reductase in Arabidopsis thaliana.* Nat Commun, 2014. **5**: p. 4617.

177. Chao, D.Y., et al., *Genome-wide association mapping identifies a new arsenate reductase enzyme critical for limiting arsenic accumulation in plants.* PLoS Biol, 2014. **12**(12): p. e1002009.

178. Styblo, M., et al., *Comparative toxicity of trivalent and pentavalent inorganic and methylated arsenicals in rat and human cells.* Archives of Toxicology, 2000. **74**(6): p. 289-299.

179. Marin, A.R., P.H. Masscheleyn, and W.H. Patrick, *The Influence of Chemical Form and Concentration of Arsenic on Rice Growth and Tissue Arsenic Concentration.* Plant and Soil, 1992. **139**(2): p. 175-183.

180. Rosen, B.P., *Families of arsenic transporters.* Trends Microbiol, 1999. **7**(5): p. 207-12.

181. Song, W.Y., et al., *A rice ABC transporter, OsABCC1, reduces arsenic accumulation in the grain.* Proc. Natl. Sci. U.S.A., 2014. **111**(44): p. 15699-704.

182. Song, W.Y., et al., *Arsenic tolerance in Arabidopsis is mediated by two ABCC-type phytochelatin transporters.* Proc Natl Acad Sci U S A, 2010. **107**(49): p. 21187-92.

183. Hartley-Whitaker, J., G. Ainsworth, and A.A. Meharg, *Copper- and arsenate-induced oxidative stress in Holcus lanatus L. clones with differential sensitivity.* Plant Cell Environ., 2001. **24**(7): p. 713-722.

184. Lee, K.W., et al., *Overexpression of alfalfa mitochondrial HSP23 in prokaryotic and eukaryotic model systems confers enhanced tolerance to salinity and arsenic stress.* Biotechnol. Lett., 2012. **34**(1): p. 167-174.

185. Mascher, R., et al., *Arsenate toxicity: effects on oxidative stress response molecules and enzymes in red clover plants.* Plant Sci., 2002. **163**(5): p. 961-969.

186. Stoeva, N., M. Berova, and Z. Zlatev, *Physiological response of maize to arsenic contamination.* Biologia Plant., 2003. **47**(3): p. 449-452.

187. Dixit, S. and J.G. Hering, *Comparison of arsenic(V) and arsenic(III) sorption onto iron oxide minerals: Implications for arsenic mobility.* Environmental Science & Technology, 2003. **37**(18): p. 4182-4189.

188. Li, R.Y., et al., *Mitigation of Arsenic Accumulation in Rice with Water Management and Silicon Fertilization.* Environmental Science & Technology, 2009. **43**(10): p. 3778-3783.

189. Grassi, C., et al., *Arobic rice: crop performance and water use efficiency.* Journal of Agriculture and Environment for International Development, 2009. **103**(4): p. 259-270.

190. Arao, T., et al., *Effects of Water Management on Cadmium and Arsenic Accumulation and Dimethylarsinic Acid Concentrations in Japanese Rice.* Environmental Science & Technology, 2009. **43**(24): p. 9361-9367.

191. Hu, P.J., et al., *Water management affects arsenic and cadmium accumulation in different rice cultivars.* Environmental Geochemistry and Health, 2013. **35**(6): p. 767-778.

192. Hu, P.J., et al., *Effect of water management on cadmium and arsenic accumulation by rice (Oryza sativa L.) with different metal accumulation capacities.* Journal of Soils and Sediments, 2013. **13**(5): p. 916-924.

193. Honma, T., et al., *Effects of soil amendments on arsenic and cadmium uptake by rice plants (Oryza sativa L. cv. Koshihikari) under different water management practices.* Soil Science and Plant Nutrition, 2016. **62**(4): p. 349-356.

194. Matsumoto, S., et al., *Evaluation of the effects of application of iron materials on the accumulation and speciation of arsenic in rice grain grown on uncontaminated soil with relatively high levels of arsenic.* Environmental and Experimental Botany, 2016. **125**: p. 42-51.

195. Farrow, E.M., et al., *Reducing arsenic accumulation in rice grain through iron oxide amendment.* Ecotoxicology and Environmental Safety, 2015. **118**: p. 55-61.

196. Chou, M.L., et al., *Inhibition of ethylenediaminetetraacetic acid ferric sodium salt (EDTA-Fe) and calciumperoxide (CaO2) on arsenic uptake by vegetables in arsenic-rich agricultural soil.* Journal of Geochemical Exploration, 2016. **163**: p. 19-27.

197. Tiberg, C., et al., *Immobilization of Cu and As in two contaminated soils with zero-valent iron - Long-term performance and mechanisms.* Applied Geochemistry, 2016. **67**: p. 144-152.

198. Penido, E.S., et al., *Biogeochemical impacts of silicon-rich rice residue incorporation into flooded soils: Implications for rice nutrition and cycling of arsenic.* Plant and Soil, 2016. **399**(1-2): p. 75-87.

199. Meharg, C. and A.A. Meharg, *Silicon, the silver bullet for mitigating biotic and abiotic stress, and improving grain quality, in rice?* Environmental and Experimental Botany, 2015. **120**: p. 8-17.

200. Seyfferth, A.L., et al., *Soil Incorporation of Silica-Rich Rice Husk Decreases Inorganic Arsenic in Rice Grain.* Journal of Agricultural and Food Chemistry, 2016. **64**(19): p. 3760-3766.

201. Norton, G.J., et al., *Identification of Low Inorganic and Total Grain Arsenic Rice Cultivars from Bangladesh.* Environmental Science & Technology, 2009. **43**(15): p. 6070-6075.

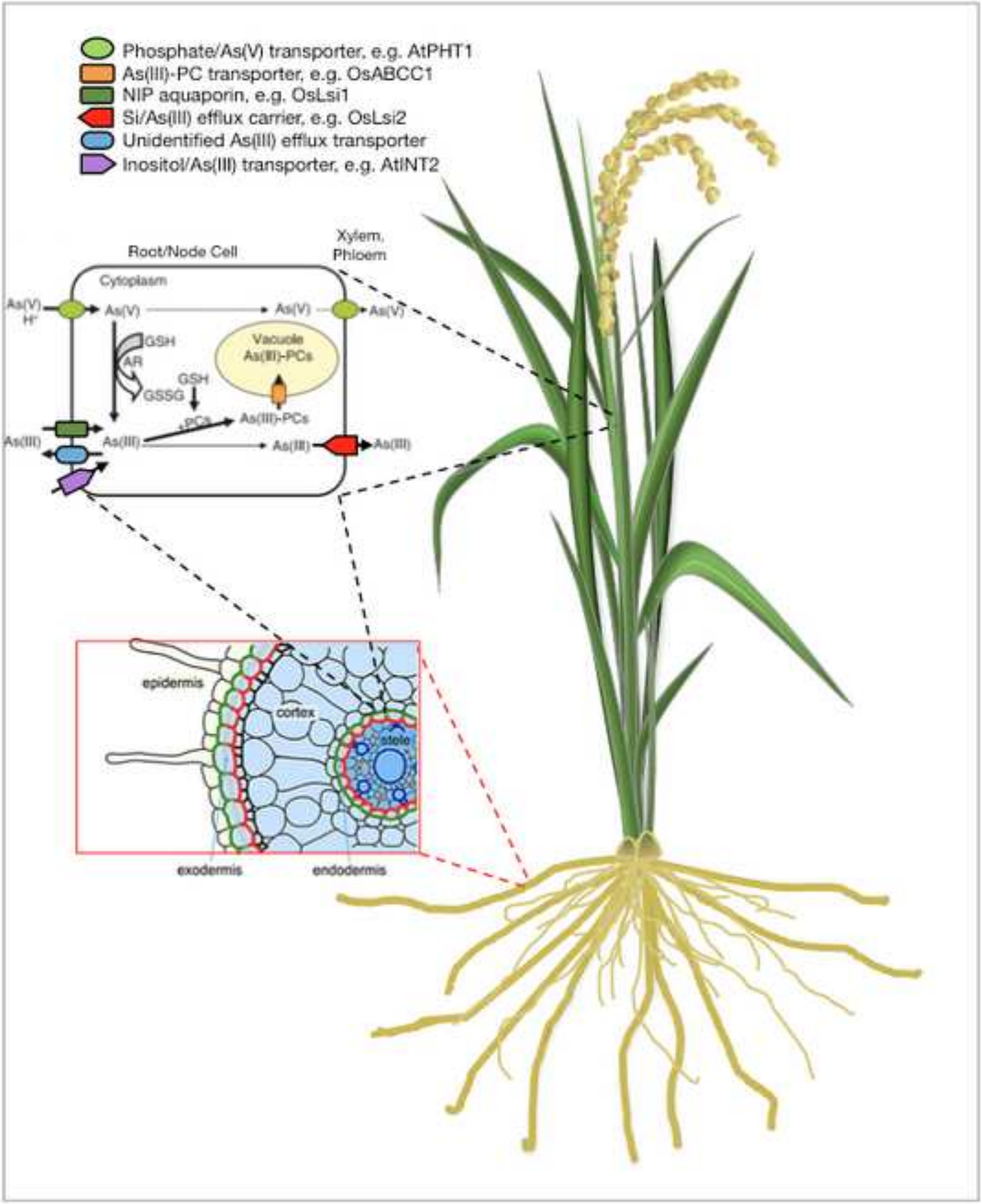
202. Duan, G., et al., *Expressing ScACR3 in rice enhanced arsenite efflux and reduced arsenic accumulation in rice grains.* Plant Cell Physiol, 2012. **53**(1): p. 154-63.

203. Song, W.Y., et al., *Arsenic tolerance in Arabidopsis is mediated by two ABCC-type phytochelatin transporters.* Proc. Natl. Acad. Sci. U.S.A., 2010. **107**: p. 21187-92.

204. Norton, G.J., et al., *Genome wide association mapping of grain arsenic, copper, molybdenum and zinc in rice (Oryza sativa L.) grown at four international field sites.* PLoS One, 2014. **9**(2): p. e89685.

205. Tang, Z., et al., *Arsenic Methylation in Arabidopsis thaliana Expressing an Algal Arsenite Methyltransferase Gene Increases Arsenic Phytotoxicity.* J Agric Food Chem, 2016. **64**(13): p. 2674-81.

Figure


Phosphate/As(V) transporter, e.g. AtPHT1
As(III)-PC transporter, e.g. OsABCC1
NIP aquaporin, e.g. OsLsi1
Si/As(III) efflux carrier, e.g. OsLsi2
Unidentified As(III) efflux transporter
Inositol/As(III) transporter, e.g. AtINT2
Root/Node Cell
Xylem, Phloem
Cytoplasm
As(V)
H+
As(V)
As(V)
As(V)
GSH
AR
GSSG
GSH
Vacuole
As(III)-PCs
+PCs
As(III)-PCs
As(III)
As(III)
As(III)
As(III)
epidermis
cortex
stele
exodermis
endodermis