

## Jury Member Report – Doctor of Philosophy thesis.

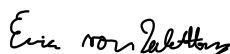
**Name of Candidate:** Alina Chernova

**PhD Program:** Life Sciences

**Title of Thesis:** Integrating high-throughput genotyping and lipidomic profiling for discovery of genetic determinants of cultivated sunflower seed oil content

**Supervisor:** Professor Philipp Khaitovich

### Name of the Reviewer:

I confirm the absence of any conflict of interest  No conflict of interest	<b>Signature:</b>    <b>Date: 28-12-2020</b>
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*The purpose of this report is to obtain an independent review from the members of PhD defense Jury before the thesis defense. The members of PhD defense Jury are asked to submit signed copy of the report at least 30 days prior the thesis defense. The Reviewers are asked to bring a copy of the completed report to the thesis defense and to discuss the contents of each report with each other before the thesis defense.*

*If the reviewers have any queries about the thesis which they wish to raise in advance, please contact the Chair of the Jury.*

### Reviewer's Report

Reviewers report should contain the following items:

- Brief evaluation of the thesis quality and overall structure of the dissertation.
- The relevance of the topic of dissertation work to its actual content
- The relevance of the methods used in the dissertation
- The scientific significance of the results obtained and their compliance with the international level and current state of the art
- The relevance of the obtained results to applications (if applicable)
- The quality of publications

The summary of issues to be addressed before/during the thesis defense

In her doctoral thesis, Chernova has used a combination of high-throughput genotyping and lipidomic phenotyping to characterize variation in seed oil content in three Russian collections of sunflowers, *Helianthus annuus*. Sunflower is an important oilseed crop, well adapted to a range of climates in Russia. She has found several new markers associated with seed oil content and quality, as well as other agronomic traits. This is an important piece of work that will facilitate harnessing the diversity of sunflower collections in Russia, and that will extend to other oilseed crops and well beyond Russia. It is an important step towards a productive scientific career for Chernova and should be congratulated. The publications are of high quality in internationally regarded journals. The topic matches the content, and cutting-edge contemporary methods are employed effectively.

The thesis is clearly written with carefully presented results. My comments on the doctoral thesis primarily focus on a few overarching themes to help improve its framing in the context of larger issues and themes. All of these are intended to help bring the excellent work to a larger international audience.

The first overarching theme that Chernova briefly addresses in her introduction (page 12) is that with population growth and climate change, immense increases in food production are required. This is a common theme in the contemporary crop genetics literature. It serves a motivation and rationale for characterizing germplasm collections to find adaptive alleles. As common as this trope has become in the literature, I believe it requires some careful examination. Although I think it is still a good motivation, we should not lose sight of the larger dynamics of the food system. Outside of Russia, incredible amounts of food are wasted, particularly in the West. Considerable strides could be made in improving food access and security were 25-40% of food not wasted in the US, Western Europe, and countries with similar food distribution systems due to spoilage. In many market economies, food production is also highly skewed towards production of animal products, which can require 10 calories of grain to provide a single calorie of meat. Furthermore, widespread income inequality means that a few have disproportionate access to food. If our aim is to reduce hunger, efforts on any of these three factors are likely to have a far larger and much more immediate impact than raising productivity of any crop. The scope of expected hunger with climate change may still exceed these steps, so raising yields is still imperative. But even with raising crop yields, genetic improvements will likely only account for a quarter of potential for improved yields. Improved agronomic management may do far more to bridge the gap between yields obtained on research stations and those achieved on actual farmers. All of this is not a reason to not also invest in crop genetics, but it is important context. We should keep in mind the relative contribution of different actions, and appreciate that genetics is just one tool. Likely improvements in crop genetics are more important for adaptation to climate change and its associated impacts (new diseases and pests, shifting patterns of abiotic stress). There is some broader historical context for this in a recent book called "The Prophet and the Wizard" (Charles Mann, 2018) that looks at the worldviews that go into how we conceptualize scientific responses to hunger and resource scarcity.

A second "big-picture" question is about the biological constraints on oil seed production. Why is it that ~60% oil content is the limit of seed oil content? What factors impact natural variation in seed oil content? How do these patterns help guide our efforts to find natural variants in seed oil content that may be agronomically useful? There is a rich ecological/evolutionary literature to be accessed on these

topics. Some of this literature is on “economic spectra” in plant traits. For species like sunflowers, where wild relatives in *Helianthus* and many other taxa in the Compositae family have wind-dispersed seeds, there is a trade-off between dispersal capacity of seeds by wind and the likelihood of germination. Larger seeds are more dispersive, but may individually have a lower germinate rate. In cultivated sunflowers this trait has been lost in domestication, but still has an evolutionary legacy. Oil composition may be shaped for selection on dispersal. A second important aspect of selection on seed fatty acid content is likely temperature. There are clear latitudinal gradients in seed fatty acid content that track background temperature. An older review from Randy Linder (Linder, C.R., 2000. Adaptive evolution of seed oils in plants: accounting for the biogeographic distribution of saturated and unsaturated fatty acids in seed oils. *The American Naturalist*, 156(4), pp.442-458.) is a good starting point for this literature, as a meeting point between agricultural and natural systems.

Understanding natural selection on fatty acid content and ratios can help design approaches to find naturally occurring variants with particular compositions in germplasm accessions. Some frameworks already exist for such searches, such as FIGS (Focused Identification of Germplasm Strategy) and Gap Analyses already exist, or genotype-environment association tests. But these methods are relatively crude, leaving open the possibility of deploying more precise tools to associate climatic conditions in particular environments with genetic variants leading to unique fatty acid profiles. I would be intrigued to see the data collected by Chernova analyzed by some of these tools, such as BayEnv, Bedassle, or Gradient Forest analysis. I suspect these general tools could be adapted to be more precise for looking for oil seed content by adding in formation on environment matching of oil seed content to different ecologies and thermal regimes. Although human impacts on distribution of landraces and cultivars may make associations of environment and genotype less precise, they may still provide insight when paired with predictions about oil seed content.

A third theme, of narrower scope, is that of minor fatty acid components. I think the discussion of this topic, which is important for the production of specialty oils, deserves more context. What biological role might these FA constituents have? What processes would lead to their low abundance in seeds or other tissues? What are the likely limits on selection in these fatty acids? Would a seed be viable with arachidic or behenic acids as a major constituent? How would its properties change?

Cytoplasmic Male Sterility has proven a valuable and durable breeding tool in sunflowers. Is there any evidence of CMS breaking down in sunflower? In maize, another crop where CMS was once widely deployed, a disease susceptibility locus (to southern blight) was closely linked to the CMS locus. Is there anything similar in sunflowers? If there is CMS breakdown, or deleterious alleles linked, then finding new CMS genes may be particularly useful. In other crops (pigeonpeas, *Cajanus cajan*, for example), many new CMS loci have come from wild relatives? Would an examination of more divergent *Helianthus* species from the secondary gene pool be helpful in this regard?

**Provisional Recommendation**

I recommend that the candidate should defend the thesis by means of a formal thesis defense

I recommend that the candidate should defend the thesis by means of a formal thesis defense only after appropriate changes would be introduced in candidate's thesis according to the recommendations of the present report

The thesis is not acceptable and I recommend that the candidate be exempt from the formal thesis defense