6. Global sensitivity analysis of a crop frost tolerance model

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6.1 Introduction

Overwintering (the capacity for plants to survive during winter) ensures autumn-sown crops the possibility to grow and produce yield in the spring-summer season. Overwintering depends on the interaction of several factors, among which the most relevant are genotype frost tolerance (Fowler et al., 2014), sowing date, frost intensity, and freezing duration (Gusta and Fowler, 1977). The last two factors determine the temperature at which crop tissues are exposed. Furthermore, for cereals soil temperature in the crown region is influenced by snow depth (Aase and Siddoway, 1979) and management practices such as soil tillage (Larsen et al., 1988). Predicting crop frost tolerance enables to estimate crop winter survival and to identify the sowing date that, in a given site, ensures the greatest chance of overwintering. This is particularly important in cold regions, where the risk of winterkill is higher than in temperate climates. While crop establishment during autumn and the subsequent overwintering make autumn-sown cereal cultivation more profitable than spring planting, under very cold winter conditions wheat is planted in spring rather than in autumn to avoid the risk of winterkill (Fowler et al., 2014).

Assessing the likelihood of crop overwintering or winterkilling in a given region is also important for cover crops. Cover crops are cultivated for their agro-ecological benefits (Justes, 2017; Tadiello et al., 2022), like reduction of soil erosion and nitrate leaching, competition with weeds, and increase of soil organic matter. In the case of winter-hardy cover crops (i.e. cover crops that successfully overwinter), the termination in the spring needs to be carried out by chemical or mechanical methods, which implies additional costs, while this is not necessary for winter-killed cover crops (Labreuche and Bodilis, 2010). Therefore, winterkill has substantial advantages for cover crops.

The acquisition and maintenance of frost tolerance depends on a complex genetic regulation, whose main drivers are environmental temperature and crop development stage. The changes involved in crop adaptation to low temperatures are dependent on several complex interactions (occurring between genotype, environment, and management) that still present knowledge gaps (Byrns et al., 2020). Within this framework, a simulation model of crop frost tolerance provides insights to integrate current scientific knowledge and to define management decisions. Comparing model outputs with field measurements allows to verify if the knowledge embedded in the model provides reliable results and therefore can be used in scenario comparisons. After the model is validated, management scenarios, obtained through site-specific simulations for several combinations of crop genotype and sowing date, could be interactively investigated by running the model with different input combinations, each corresponding to a different scenario. This allows the evaluation of production risks related to frost, as well as of breeding and agronomic strategies to maximize production potential of cash crop cultivation. On the other hand, what-if scenarios could be investigated to optimize autumn-winter cover crop management to increase or avoid winterkill occurrence (depending on the cropping system). Furthermore, these evaluations could be applied to assess the possible effects of climate change on crop overwintering or winterkill occurrence.

Several frost tolerance simulation models are available in literature. The most important ones were developed for winter wheat; these are the model by Byrns et al. (2020), FROSTOL (Bergjord et al., 2008) and the model by Lecomte et al. (2003). The latter estimates crop frost resistance on day d (R_d , °C), which is defined as the temperature below which the first leaf damage occurs, using air temperature as driving variable. FROSTOL and Byrns et al. (2020) simulate frost tolerance as the "lethal temperature 50" (LT_{50} , °C), which is defined as the soil temperature in the crown region at which 50% of the plants are killed in an artificial freeze test (Bergjord et al., 2008). In all these models, frost tolerance is increased by low temperature hardening (that decreases the frost tolerance temperature) and is lowered by de-hardening (that increases frost tolerance temperature). Both FROSTOL and the model by Byrns et al. (2020) decrease frost tolerance due to two types of stress: respiration under a snow cover and exposure to sub-lethal temperature. The model by Byrns et al. (2020) is the most recent version of the modelling approach developed by Fowler et al. (1999), that was also adopted by Bergjord et al. (2008) in the FROSTOL model.

All these models use several parameters that need to be calibrated by adjusting their value to obtain the best possible fit between simulated and measured values. Calibrating many parameters requires extremely large data sets and can lead to the identification of local instead of optimal combinations of parameter values. Therefore, even in complex agronomic models, only few relevant parameters should be calibrated. These can be identified using sensitivity analysis, a statistical technique that assigns model output variability to different model input parameters (Saltelli, 2004). Sensitivity analysis is therefore needed for a more informed use of a model. It can be performed using local methods such as the one-factor-at-a-time (OAT) screening techniques, or global methods such as the one designed by Morris (1991) or the one developed by Sobol (Saltelli et al., 2010). Since local sensitivity analysis methods are performed by varying one parameter at time, they do not describe parameter interactions that are instead investigated by global methods, performed by varying simultaneously all model parameters. Therefore, global methods allow a more profound understanding of model behavior. To the best of our knowledge, there are no sensitivity analyses in the literature regarding the model by Byrns et al. (2020) yet. Model output sensitivity to parameters is strongly influenced by environmental conditions, like weather, soil and management. For this reason, since soil temperature is the main environmental driver of the model, different sites and sowing dates need to be considered in the sensitivity analysis.

Therefore, the objective of this paper is to perform a global sensitivity analysis of the frost tolerance model by Byrns et al. (2020) applied to winter crops (cereals and cover crops), assessing parameter rankings for different sowing dates and sites.

6.2 Materials and methods

6.2.1 Model description

The crop winter survival model (Byrns et al., 2020) expresses frost tolerance as the "lethal temperature 50" (LT_{50} , °C). The LT_{50} is a state variable calculated daily using as a driving variable the average daily soil temperature in the crown region (T_c , °C). Its rate variable (ΔLT_{50} , °C d ⁻¹, Eq. 1) describes four different processes involved in the gain or loss of frost tolerance: low temperature acclimation (hardening, $\Delta LT_{50H flow}$, Eq. 3, and $\Delta LT_{50H rate}$, Eq. 9), dehardening ($\Delta LT_{50D flow}$, Eq. 4, and $\Delta LT_{50D rate}$, Eq. 11) and loss of frost tolerance due to snow cover ($\Delta LT_{50R flow}$, Eq. 5) or to sub-lethal temperature stress ($\Delta LT_{50S flow}$, Eq. 6). Even if model

equations are clearly explained in Byrns et al. (2020), as well as their experimental derivation, they are reported also here to better understand the effect of model parameters on the simulated outputs. Model equations are listed below, while parameters and variables are explained in Table 1 and Table 2, respectively. Hardening and dehardening are influenced by the threshold induction temperature of the crop (Eq. 2) and by crop development stage, that is represented by the progress to the vegetative/reproductive transition (Eq. 12 and 13). The progress to the transition is simulated through the fulfilling of: a photoperiod requirement (Eq. 14, 15 and 16), a vernalization requirement (Eq. 17, 18 and 19) and a minimum leaf number requirement (Eq. 20 and 21). The loss of frost tolerance due to sub-lethal temperature exposure is influenced by the minimum value of LT_{50} reached during the simulation (Eq. 7) and by the accumulated dehardening amount (Eq. 8). Respiration under a snow cover stress is simulated according to the average (T_m , °C) soil temperature of the previous 10 days (ranging between *RESP_Tmin* and *RESP_Tmax*) and its standard deviation (T_{sd} , °C, being lower than *RESP_Tsd*).

$$\frac{\Delta LT_{50}}{\Delta t} = -\Delta LT_{50H\,flow} + \Delta LT_{50D\,flow} + \Delta LT_{50R\,flow} + \Delta LT_{50S\,flow}$$
[1]

$$T_i = 3.72135 - 0.401124 \times LT50c$$
^[2]

$$\Delta LT_{50H flow} = \begin{cases} 0 & if \Delta LT_{50R flow} > 0\\ \Delta LT_{50H rate} \times VRT_{factor} & if \Delta LT_{50S flow} = 0\\ 0 & otherwise \end{cases}$$
[3]

$$\Delta LT_{50D flow} = \begin{cases} \Delta LT_{50D rate} & if T_c > T_i \text{ and } LT_{50} < LT50i \\ \Delta LT_{50D rate} \times (1 - VRT_{factor}) & if T_c > LT50i \text{ and } LT_{50} < LT50i \\ 0 & if \Delta LT_{50R flow} > 0 \text{ or else} \end{cases}$$

$$[4]$$

 $\Delta LT_{50R flow} =$

$$\begin{cases} \frac{RESP1 \times (e^{RESP2 + RESP3 \times T_c - RESP4})}{RESP5} & IF (T_m < RESP_T max AND T_m > RESP_T min AND T_{sd} < RESP_T sd) \\ 0 & otherwise \end{cases}$$
[5]

$$\Delta LT_{50S\,flow} = \begin{cases} \left| \frac{LT_{50min} - T_c}{e^{LOWT_1 \times (LT_{50min} - T_c) - LOWT_2}} \right| & \text{if } LT_{50} < T_c < LT_{50min} \\ and \ LT_{50} - dehardAmtStress < LT50i \text{ and } T_c < LT50i \\ otherwise \end{cases}$$
[6]

$$LT_{50min\ flow} = \begin{cases} LT_{50} - LT_{50min} & \text{if } LT_{50} < LT_{50min} \\ 0 & \text{else} \end{cases}$$
[7]

$$\frac{\Delta dehardAmtStress}{\Delta t} = -\left(\Delta LT_{50R\,flow} + \Delta LT_{50S\,flow}\right)$$
[8]

$$\Delta LT_{50H\,rate} = max \left(0; \, HARDrate \, \times (T_i - T_c) \times \left(LT_{50} - LT_{50adj}\right)\right)$$
[9]

$$LT_{50adj} = LT50c - dehardAmtStress$$
[10]

$$\Delta LT_{50D \ rate} = \frac{DEHARD1}{1 + e^{DEHARD2 - DEHARD3 \times min\left(T_c, \ T_i\right)}}$$
[11]

$$VRT_{progress} = min (min(min(1; mflnFraction); photoProg); vernSaturation)$$
[12]

$$VRT_{factor} = \frac{1}{1 + e^{VRT_{fct1}(VRT_{progress} - VRT_{fct2})}}$$
[13]

$$photoProg = \begin{cases} min \ (photoReqFraction, 1) & if \ PHOTOcoeff > 0 \\ 0 & else \end{cases}$$
[14]

$$photoFactor = \begin{cases} \left| \frac{PHOTO1}{\left[1 + e^{PHOTO2 \times (DL - PHOTOcrit) - PHOTO3 \times (T_c - PHOTO4)}\right] - PHOTO1} \right| & if T_c > 0 \text{ and } \Delta LT_{50R flow} = 0 \text{ [15]}\\ 0 & else \end{cases}$$

$$photo_{flow} = \frac{photoFactor}{PHOTO5 \times PHOTOcoeff}$$
[16]

$$vernSaturation = \begin{cases} \min\left(\frac{vernDays}{VERNreq}, 1\right) & if VERNreq > 0\\ 1 & else \end{cases}$$
[17]

$$delta = Log(2)/Log((VERN_Tmax - VERN_Tmin)/(VERN_Topt - VERN_Tmin))$$
[18]

vernRate =

$$\begin{cases} 1 & if VERN_Tmin \le T_c \le VERN_Topt \\ (2 \times (T_c - VERN_Tmin)^{delta} \times (VERN_Topt - VERN_Tmin)^{delta} - (T_c - VERN_Tmin)^{2delta})/(VERN_Topt - VERN_Tmin)^{2delta} \\ if VERN_Topt < T_c \le VERN_Tmax \\ 0 & else \end{cases}$$

$$[19]$$

 $DDReqCurrentTemp = max (GDDmin, (GDD1 \times GDDmin - GDD2) \times (T_c - GDD3) + GDDmin)$ [20]

$$mfln_{flow} = \frac{\max(T_c, \ 0)}{DDReqCurrentTemp}$$
[21]

$$mflnFraction_t = mflnFraction_{t-1} + mfln_{flow}$$
^[22]

The rate equations are used to integrate the state variable with the Euler method with a daily time step. Compared to the original implementation, we have restricted the range of possible values of the two stress rate variables (Eq. 5 and 6), with the purpose of increasing model stability. The computer code was tested for correctness against the original implementation reported in Byrns et al. (2020).

6.2.2 Parameter ranges and distributions

The model by Byrns et al. (2020) employs 36 parameters that we divided in two categories, explicit and implicit parameters (Table 1). Explicit parameters are the ones with a clear biological meaning (identified by a name in the original publication), while implicit parameters are empirical coefficients indicated as numbers in original model equations. There are 13 explicit parameters: they mainly describe temperature values (*LT50i*, *LT50c*, vernalisation temperature range, stress occurrence temperature range), but also vernalisation and photoperiodic requirements (*GDDmin*, *VERNreq*, *PHOTOcrit*).

The default values of all parameters, reported in Table 1, were derived from the original model implementation (Byrns et al., 2020) dedicated to several winter or spring cereals (wheat, rye, barley, and oat). Then, lacking detailed information on parameter values in the scientific literature, the range of variation for each parameter was obtained by adding and subtracting the 30% of the default value to the default value itself (Esprey et al. 2004; Song et al., 2012). Afterwards, some range limits were adapted to include in the analysis also winter cover crop species that experience frost damage at warmer temperatures compared to cereals, i.e. near 0 °C. The upper limit for *LT50c* was set at -3.89 °C (Clark, 2007). For the initial value of *LT50* (*LT50i*), the upper limit was fixed to -0.805 °C (according to Bergjord et al., 2008), and the lower limit to -3.9 °C. To ensure a range of variation of *PHOTOcrit* able to encompass cereals and cover crops, we fixed the lower limit to the default value of a white mustard cover crop (10 h) decreased by 30%, while the upper limit was obtained using a default value for winter wheat cultivars (13.5 h) increased by 30%. The statistical distribution of all model parameters (both explicit and implicit) was considered uniform, since there is limited information in the literature regarding the distribution of these quantities.

6.2.3 Modelling scenarios of the sensitivity analysis

Sensitivity analyses were carried out for each combination of three sowing dates, three sites, and 20 years (autumn 1999 - spring 2020) that were chosen to sample the variability of European climates and sowing dates. The selected sowing dates (SD) were September 1st (SD1), October 1st (SD2) and October 30th (SD3). The selected sites were Sant'Angelo Lodigiano (Latitude 45.26°N, Longitude 9.38°E, Northern Italy),

Merzenich (Latitude 50.82°N, Longitude 6.55°E, North Rhine-Westphalia, Germany) and Karklupėnai (Latitude 54.58°N, Longitude 22.77°E, Pajevonys Eldership, Lithuania). The Köppen climate classification of the sites are: Cfa (humid subtropical; Sant'Angelo Lodigiano), Cfb (oceanic; Merzenich) and Dfb (warm summer continental; Karklupėnai). The daily soil temperature at the crown level needed as input for the model was simulated with the ARMOSA cropping system model (Perego et al., 2013); these simulations were run three times in each site, for a 20-year monocropping, using SD1, SD2 and SD3 as sowing dates. Each simulation was ended on April 30th of the year after sowing. Soil inputs (texture and organic carbon concentration) used by ARMOSA were derived from a regional soil map (ERSAL, 2000) for Sant'Angelo Lodigiano, and from the European database LUCAS (Jones et al., 2020) for the other two sites. Soil descriptions of the three sites are reported in Table 3. Weather inputs were obtained from the regional agency for environment protection (ARPA) weather station network for the Italian site, while for the other two sites they were derived from the gridded meteorological database Agri4Cast (Biavetti et al., 2014). A summary of the soil temperature simulated by ARMOSA is reported in Table 4.

The selected model outputs for sensitivity analysis were: the minimum *LT*₅₀ reached during the simulation and the days required to reach it (exemplified in Fig. 1). These quantities were evaluated at the end of the period comprised between sowing and February 28, since from a preliminary study it emerged that, especially for the warmer site (Sant'Angelo Lodigiano, Italy), frost tolerance differences due to sowing date (SD1, SD2, and SD3) can be observed only after winter and not during autumn.

Furthermore, the effect of the inter-annual weather variability on the main model output (weekly minimum LT_{50}) variations, caused by parameter values modifications induced during the sensitivity analysis procedure, was evaluated for each site using two reference autumn-winter seasons. The two reference seasons, a cold and a warm season, were selected between the 20 years of meteorological inputs to represent weather variability in each site. These seasons were selected, for each site, on the basis of the monthly minimum and maximum average daily soil temperature, and of the number of days during which the soil temperature was negative. A season was selected, between the 20 available seasons, as cold when it minimized both soil

temperatures and it maximized the number freezing days, while it was selected as warm when it maximized the soil temperatures, and it minimized the freezing days.



Figure 1. Selected model outputs for sensitivity analysis: minimum LT₅₀ value during the simulation and days to reach it.

After having examined the results of the first sensitivity analysis, we realized that LT50c was by far the most important parameter, while the sensitivity of the other parameters was much smaller. Therefore, to better investigate the role of the other model parameters, additional sensitivity analyses (resulting from the same combinations of three sowing dates, three sites, and 20 years) were carried out by adopting three different fixed (instead of varying) values of the parameter *LT50c*. The three *LT50c* values were chosen to represent the maximum frost tolerance of three plant species that are commonly used as autumn-winter cover crops: *Secale cereale* L. (rye), *Avena sativa* L. (oat), and *Sinapis alba* L. (white mustard). The average reference values retrieved from various literature sources (Byrns et al., 2020; Clark, 2007) were rounded to the nearest integer value: -29, -8 and -4 °C for winter rye, oat, and white mustard, respectively.

6.2.4 Sobol method for global sensitivity analysis

The Sobol method for global sensitivity analysis is a variance-based method that performs a decomposition of the total model output variance (V) in partial variances that are caused by a single parameter i (V_i) and by the interaction between two (V_{ij}) or more (V_{ijm}, ...) parameters (Saltelli et al., 2010). Considering windependent model parameters, the decomposition of the total variance is executed as it follows:

$$V = \sum_{i} V_{i} + \sum_{i < j} V_{ij} + \sum_{i < j < m} V_{ijm} + \dots + V_{12\dots w}$$

The ratios between partial variances and total variance represent the sensitivity indices, whose values represent the portion of total variance that is caused by the variation of single parameters or by their interaction. The first-order sensitivity index ($S_i = V_i/V$) measures the additive effect of the parameter *i* on the model output. The second-order sensitivity index ($S_{ij} = V_{ij}/V$) measures the effect of the interaction between the parameters *i* and *j* on the model output. The effect of the interaction between parameters can also be accounted for by using a total-order index (ST_i): the difference between ST_i and S_i is the measure of the total model output variance due to the all the interactions between the parameter *i* and all the other model parameters. The sensitivity analysis was performed by estimating first, second and total-order sensitivity indices according to the implementation proposed in Saltelli et al. (2010). The number of quasi-Montecarlo simulation runs was 65.536. The Sobol sequence generator was initialized with the set of direction numbers provided by Joe and Kuo (2008).

6.3 Results

6.3.1 Sensitivity indices for northern Italy

For the site in northern Italy, model outputs showed high variability due to seasonal differences of weather, as reported in Figure 2. During the warmer autumn-winter season (2006/2007), the lethal temperature 50% remained in a narrow range until the end of January (week 23), then its variability due to parameter variations increased but remained lower compared to the same period of the cold season (2011/2012). In 2021/2022, model output started to show substantial variability due to parameter combinations from the beginning of October (week 7).



Figure 2. Variations of model output (weekly minimum LT_{50}) due to parameter combinations during two seasons: 2006/2007 (warm season) and 2011/2012 (cold season) for the site Sant'Angelo Lodigiano (Italy). The simulations reported in this graph were run from the first sowing date (September 1st) to April 30. The boxplots show output variability due to the different parameter combinations used to run the sensitivity analysis.

When considering the first model output, i.e. the minimum value of LT₅₀ (for the period sowing date-February

28), the most influent parameter (data not shown in figures or tables) was LT50c, that explained more than

95% of model output variance for the three sowing dates. Indeed, the first-order sensitivity coefficient (S1)

was equal to 95%, 96%, and 98% for the first (SD1), second (SD2), and third (SD3) sowing date, respectively,

indicating that this parameter was always the most relevant. The sum of S1 coefficients for all parameters was 97%, 98%, and 99% for the first, second, and third sowing date, respectively.

When considering the second model output, i.e. the days needed to reach the minimum *LT*₅₀ value, the parameters *PHOTOcrit*, *VRTfct2* and *LT50c* were the most important, as reported in Figure 3.



Figure 3. First-order (S1) and total-order (ST) sensitivity coefficients for the days to reach the minimum value of LT_{50} for the three sowing dates (SD1, SD2, and SD3) in Sant'Angelo Lodigiano (Italy). Sensitivity coefficients were estimated for the period sowing date-February 28.

PHOTOcrit and VRTfct2 effects were higher for SD1 and SD2 than for SD3, while LT50c effect was higher for

SD3. Furthermore, LT50c effect showed high variability both within the same sowing date and between the

different sowing dates. First-order sensitivity coefficient of VRTfct2 was equal to 26%, 23% and 17% for SD1,

SD2, and SD3, respectively, while for PHOTOcrit these values were equal to 20%, 19% and 15%, and for LT50c

were 6%, 8%, and 11%. The average sum of S1 sensitivity coefficients for all parameters was 61%, the same

for all sowing dates. For this second output second-order coefficients were lower than 8% (Figure 4).



Figure 4. Second-order (S2) sensitivity coefficients for the days to reach the minimum value of LT_{50} for the three sowing dates (SD1, SD2, and SD3) in Sant'Angelo Lodigiano (Italy). Sensitivity coefficients were estimated for the period sowing date-February 28.

The sensitivities obtained after fixing the value of *LT50c* for three cover crop species (white mustard, oat, and rye) are reported in Figure 5. For the highest *LT50c* values considered (-4 and -8 °C, respectively for white mustard and oat), the most important parameter for the first model output (minimum *LT₅₀* value; Figure 5A) was the initial value of *LT₅₀* (*LT50i*): for mustard, its average S1 for the three sowing dates was 62%, while for oat it was 14%. For the early-planted oat (SD1) the parameter *VRTfct2* (S1 equal to 18%) and *PHOTOcrit* (S1 equal to 13%) were also relevant. For the lowest *LT50c* value instead (-29 °C, rye) the highest S1 values were obtained by *Ti2* (S1 = 9%) for SD1, and by *VRTfct2* for SD2 (S1 = 12%) and SD3 (S1 = 27%). Interactions between parameters (indicated by high differences between ST and S1) were mainly responsible for the variations of the second output considered (days to reach the minimum *LT₅₀* value; Figure 5B) for all species and sowing dates. For all sowing dates, the most relevant parameter for mustard and oat was *VRTfct2* (S1 = 17% for mustard and 23% for oat), while for rye it was *PHOTOcrit* (S1 = 25%).



Figure 5. First-order (S1) and total-order (ST) sensitivity coefficients for the minimum value of LT_{50} (A) and for the days to reach it (B) for the combination of three sowing dates (SD1, SD2, and SD3) and three cover crop species (white mustard, oat, and rye) in Sant'Angelo Lodigiano (Italy). Sensitivity coefficients were estimated for the period sowing date-February 28.

6.3.2 Sensitivity indices for North Rhine - Westphalia (Germany)

For this site, model output variations among years (described by differences of the box length in Figure 6

between the warmer and the colder season) were low and limited to the period between the beginning of



October (week 6) and the beginning of November (week 10).

Figure 6. Variations of model output (weekly minimum LT_{50}) due to parameter combinations during two seasons: 2013/2014 (warm season) and 2010/2011 (cold season) for the site Merzenich (Germany). The simulations reported in this graph were run from the first sowing date (September 1st) to April 30. The boxplots show output variability due to the different parameter combinations used to run the sensitivity analysis.

For the first output (minimum *LT*₅₀ of the simulation) the most influential parameters were the same found in northern Italy, even with slight differences of the variability of the effects (data not shown in figures or tables). The parameter *LT50c* showed a first-order sensitivity coefficient (S1) higher than 95% (95%, 96% and 98% for SD1, SD2, and SD3, respectively). As already found in Italy, also in this site the sum of the first-order sensitivity coefficients of all parameters was higher than 97% (97%, 98% and 99% for SD1, SD2, and SD3, respectively). Regarding the second output (days to reach the minimum *LT*₅₀ value), higher first-order sensitivity coefficients (as reported in Figure 7) were registered for the parameters involved in the fulfilment of photoperiodic requirements (*PHOTOcrit* and *PHOTO4*), for the parameter *LT50c*, and for the parameters used to estimate the factor describing the progress to the vegetative/reproductive transition (*VRTfct2*). The sum of S1 sensitivity coefficients was equal to 62%, on average for the three sowing dates. Second-order coefficients were lower than 7.5% (Figure 8), with *PHOTOcrit* being the most important.



Figure 7. First-order (S1) and total-order (ST) sensitivity coefficient for the days to reach the minimum value of LT_{50} for the three sowing dates (SD1, SD2 and SD3) in Merzenich (Germany). Sensitivity coefficients were estimated for the period sowing date-February 28.



Figure 8. Second-order (S2) sensitivity coefficient for the days to reach the minimum value of LT_{50} for the three sowing dates (SD1, SD2 and SD3) in Merzenich (Germany). Sensitivity coefficients were estimated for the period sowing date-February 28.

The results of the analysis carried out by single cover crop species are reported in Figure 9. For the minimum LT_{50} value (Figure 9A), the initial value of LT_{50} (LT50i) was the most relevant parameter for mustard in all sowing dates, with an average S1 value equal to 67%. LT50i was the most relevant parameter also for SD2 and SD3 in oat, with an average S1 value of 17%, while for SD1 highest S1 (17%) was for VRTfct2. In the case of rye, the minimum LT_{50} was influenced by several parameters. For SD1 and SD2 the highest S1 value (13%) was recorded for VRTfct2, while in SD3 it was obtained by PHOTO4 and PHOTO1 (24%).



Figure 9. First-order (S1) and total-order (ST) sensitivity coefficient for the minimum value of LT_{50} (A) and for the days to reach it (B) for the combination of three sowing dates (SD1, SD2 and SD3) and three crops (white mustard, oat, and rye) in Merzenich (Germany). Sensitivity coefficients were estimated for the period sowing date-February 28.

When considering the days to reach the minimum LT_{50} value (Figure 9B), for rye the most relevant parameters in all sowing dates were *PHOTOcrit* and *VRTfct2* (S1 = 30% and 22%, respectively), while for mustard and oat it was *VRTfct2* (S1 on average equal to 20%). For all cover crop species, interactions among parameters were mainly responsible for the variations of this output for all sowing dates, as indicated by high differences between ST and S1 for most parameters.

6.3.3 Sensitivity indices for South-West Lithuania

For Lithuania, model output variability due to differences among years was low, as reported in Figure 10. Differences between model output variability during warmer and colder seasons smoothed out starting from mid-January (week 12).



Figure 10. Variations of model output (weekly minimum LT_{50}) due to parameter combinations during two seasons: 2013/2014 (warm season) and 2002/2003 (cold season) for the site Karklupėnai (Lithuania). The simulations reported in this graph were run from the first sowing date (September 1st) to April 30. The boxplots show output variability due to the different parameter combinations used to run the sensitivity analysis.

Regarding the minimum LT₅₀ reached during the simulation, the most relevant parameter (data not shown in

figures or tables) was LT50c, whose S1 and ST were characterized by lower variability for SD3 compared to

the first two sowing dates. First-order sensitivity coefficient for LT50c was 96%, 98%, and 100% for the three

sowing dates. The sum of the first-order sensitivity coefficients was higher than 98% in all the sowing dates.

The parameters having the highest effects on the days to reach the minimum *LT*₅₀ value were *LT50c*, *PHOTOcrit*, *VRTfct2*, and *RESP_Tmin* (Figure 11). Their first-order sensitivity coefficients were, on average for the three sowing dates, 14% for *LT50c*, 13% for *VRTfct2*, 10% for *PHOTOcrit*, and 6% for *RESP_Tmin*. The sum of S1 coefficients was 58% on average for the three sowing dates. Second-order coefficients were always lower than 8% (Figure 12).



Figure 11. First-order (S1) and total-order (ST) sensitivity coefficient for the days to reach the minimum value of LT₅₀ for the three sowing dates (SD1, SD2 and SD3) in Karklupenai (Lithuania). Sensitivity coefficients were estimated for the period sowing date-February 28.



Figure 12. Second-order (S2) sensitivity coefficient for the days to reach the minimum value of LT₅₀ for the three sowing dates (SD1, SD2 and SD3) in Karklupėnai (Lithuania). Sensitivity coefficients were estimated for the period sowing date-February 28.

Considering the sensitivity analysis carried out for separate cover crop species, the minimum LT_{50} (Figure 13A) for mustard was mainly affected by LT50i in all sowing dates (average S1 = 62%). For oat LT50i was the most important parameter for SD2 and SD3 (S1 = 17%), while for SD1 the most relevant parameter was VRTfct2 (S1 = 17%), followed by LT50i (S1 = 16%). For rye the highest S1 was for VRTfct2 in SD1, or Ti1 in SD2, and for HARDrate in SD3, with S1 values respectively equal to 14%, 19%, and 18%. For late-planted rye (SD3) the standard deviations of S1 were higher compared to early-planted rye (SD1 and SD2) and to the other crops. The variations of the days to reach the minimum LT_{50} value (Figure 13B) were predominantly caused by interactions among parameters, especially for SD1. For SD3 the most relevant parameters were LT50i for mustard (S1 = 27%), and $RESP_Tmin$ for oat (S1 = 7%) and rye (S1 = 25%). For rye, the most important parameters were VRTfct2 (S1 = 26%) and PHOTOcrit (S1 = 23%) in SD1, while in SD2 the same parameters both obtained a S1 value equal to 17%.



Figure 13. First-order (S1) and total-order (ST) sensitivity coefficient for the minimum value of LT_{50} (A) and for the days to reach it (B) for the combination of three sowing dates (SD1, SD2 and SD3) and three crops (white mustard, oat, and rye) in Karklupenai (Lithuania). Sensitivity coefficients were estimated for the period sowing date-February 28.

6.4 Discussion

6.4.1 Seasonal differences

Differences in model output between colder and warmer seasons within a site were higher for the Italian site with a warmer climate (Sant'Angelo Lodigiano) than for the two sites with a colder climate (Merzenich and Karklupėnai). Therefore, we hypothesize that the degree of frost tolerance that can be expressed by a crop is expected to be more season-dependent in warmer climates. This is relevant for the evaluation of winterkill chance of winter cover crops, which likely will be more variable among years in warmer sites. Inter-annual variability of frost damage occurrence and extent is also reported for winter wheat cultivation in sites with temperate climate (Lecomte et al., 2003), where winterkill events are less frequent than in continental climate sites.

6.4.2 Sensitivity of minimum LT₅₀ to model parameters and sowing dates

For the first model output (minimum LT_{50}) the sum of first-order sensitivity coefficients was higher than 95% for all the combinations of sowing dates and sites, mainly due to the very important contribution of LT50c (maximum frost tolerance of the cultivar). This implies that a strong dependence exists between this parameter and the minimum LT_{50} of the simulation, and that interactions among parameters are almost irrelevant for this output. This is justified by the fact that the interactions between LT50c and other parameters are always lower than the 4%. This result is consistent with the relevance showed by the lowest temperature tolerance (CTMX, °C) in the outcomes of a sensitivity analysis applied to an alfalfa (*Medicago sativa* L.) yield model (Kanneganti et al., 1998).

Differences of first-order sensitivity (S1) to *LT50c* among sowing dates were higher in colder sites (Merzenich and Karklupėnai) than in the warmer site (Sant'Angelo Lodigiano). The sensitivity to *LT50c* was always higher for SD3 than for the earlier sowing dates (SD1 and SD2), since in this case the shorter time frame comprised between the sowing date and February 28 narrows the days during which hardening process takes place. This effect was intensified in the colder sites, where the incidence of this parameter was higher than in the warmer site, since soil temperature was more frequently lower than the threshold required for acclimation.

6.4.3 Sensitivity of the number of days needed to reach the minimum LT₅₀ to model parameters and sowing dates

For the days to reach the minimum LT_{50} the sum of first-order sensitivity coefficients for all the combinations of sowing dates and sites, was on average equal to 60%; the parameters contributing most were *VRTfct2*, *PHOTOcrit* and *LT50c* (average S1, across all site*sowing date combinations, respectively equal to 19, 16 and 12%). Second-order sensitivity coefficients for this output were always lower than 8%, indicating that the number of days to reach the minimum LT_{50} value strongly depended not only on a few very important parameters and on interactions among two of them, but also on interactions among three or more parameters (32%). This result was likely due to the method employed to simulate crop development, that requires the fulfillment of three requirements (vernalization, photoperiod and minimum leaf number), thus making the parameters involved in these algorithms interact with each other. The effect of crop development parameters on the days required to reach the minimum LT_{50} value is higher than their effect on the minimum LT_{50} value. These parameters strongly influence the date of the end of the hardening process (that corresponds to the vegetative/reproductive transition date), but their effect on the minimum LT_{50} value is almost negligible since when the transition occurs the LT_{50} value is frequently close to the minimum value that can be assumed by this variable (LT50c). Therefore, these parameters produce small variation of the LT_{50} minimum value.

In all sites, the parameters involved in the simulation of the transition to the reproductive phase (among which the most influential are *PHOTOcrit* and *VRTfct2*) had a lower first-order sensitivity coefficient for SD3 compared to SD1 and SD2. Those two parameters were less influent in SD3 because the requirements for the vegetative/reproductive transition were fulfilled late, mainly after the minimum LT_{50} value was already reached. The transition to the reproductive phase slows down (and potentially almost stops) the decrease of LT_{50} : in the case of late sowing dates the transition is delayed by environmental limiting factors (temperature and photoperiod). Therefore, in the SD3 scenarios, because the transition was delayed, the parameters involved in its simulation did not have the opportunity to substantially influence the number of days required to reach the minimum LT_{50} value. The parameter *PHOTOcrit* (Eq. 15) is used to estimate the factor describing the progress to photoperiod requirement saturation, while the parameter *VRTfct2* (Eq. 13) is involved in the calculation of the factor simulating the vegetative/reproductive transition.

Since the parameter *PHOTOcrit* was varying between 7.0 and 17.5 h, it had little effect on the model output for late sowing dates in the site located at the highest latitude (Karklupenai) where daylength shortens quickly from the beginning of October, reaching values of 9 h at the end of the month. This situation, together with a slow accumulation of thermal units, reduced the sensitivity also of the parameter *VRTfct2*.

6.4.4 Conclusions for model calibration

We summarize here the parameters that should be modified when this frost model is adapted to a given species and variety. The parameter to which the main model output (minimum LT_{50} value) is most sensitive for all the combinations of site and sowing date is *LT50c*, that expresses the maximum frost tolerance of the cultivar (i.e. the minimum LT_{50} that can be reached during the simulation). Other parameters that should be

calibrated, both in colder and warmer sites as well as for early and late sowing dates, are the ones involved in the simulation of crop development stage (*VRTfct2* and *PHOTOcrit*) that influence the other main model output considered (days to reach the minimum LT_{50} value). The fact that these two parameters were less relevant in the late sowing date scenarios (SD2 and SD3) indicates that, to correctly consider the effect of sowing date and crop development on frost tolerance acquisition, these parameters need to be carefully calibrated. This should avoid simulating the reproductive transition beforehand, thus overestimating LT_{50} and therefore underestimating the frost tolerance of late-planted crops. At the same time, it should protect the model user from the opposite simulation error. On the contrary, the other photoperiodic parameters were not relevant (S1 < 5%).

The parameters involved in the loss of frost tolerance due to the respiration under a snow cover (Eq. 5) showed variable importance depending primarily on the site, and then on the sowing date. In the Italian and German sites, this process had only limited relevance for the third sowing date (and no relevance for SD1 and SD2), therefore efforts to calibrate them can be avoided. On the contrary, for the Lithuanian site several of these parameters showed a high total-order effect combined with low first-order effect, meaning that the interactions of these parameters with others were relevant. Therefore, in colder climates, where the more frequent presence of persistent snow cover leads often to the loss of frost tolerance, the calibration of the following parameters is required: RESPdays, RESP_Tsd, RESP4, RESP2, and RESP_Tmin.

The results obtained with fixed LT50c values, representing specific crop frost tolerance potential, indicate that for less frost-resistant crops (white mustard and oat) the site and sowing date effects on output sensitivity are less relevant compared to more frost-resistant crops. Parameter rankings for mustard and oat (SD1, SD2, and SD3) are more consistent among sites than the rankings obtained for rye. Therefore, the calibration of the most relevant parameters for less frost resistant crops could require less extended calibration datasets.

6.5 Conclusions

We reported the first sensitivity analysis study of the model by Byrns et al. (2020), performed by applying the model in different scenarios involving three sowing dates and three climates (Cfa, Cfb and Dfb). Each combination of the above-mentioned factors was tested for 20 years. Differences in model output between colder and warmer seasons were higher in the site with warmer climate than in the sites with colder climate. In all the simulated scenarios (sowing date x site combinations) the minimum *LT*₅₀ was primarily depending on the crop potential frost tolerance parameter (*LT50c*), while the number of days needed to reach the minimum *LT*₅₀ value depended strongly on higher grade parameter interactions. These interactions involved the parameters governing crop development (*PHOTOcrit*, *VRTfct2*), and limited to the coldest site, the parameters for the simulation of crop respiration under a snow cover (*RESPdays*, *RESP_Tsd*, *RESP4*, *RESP2*, and *RESP_Tmin*).

Tables

Table 1. Parameters of the model by Byrns et al. (2020). Parameters are called 'explicit' when they are explicitly defined by the authors, and 'implicit' when they are empirical numerical coefficients in model equations (without a name given by model authors).

Category	Label	Unit	Definition Default		Lower limit	Upper limit
explicit parameter	LT50i	°C	LT ₅₀ initial value	-3.00	-3.90	-0.81
explicit parameter	LT50c	°C	maximum frost -24.00 tolerance of the cultivar (minimum LT ₅₀ value)		-31.20	-3.89
explicit parameter	GDDmin	unitless	growing degree days after planting to produce the minimum number of leaves on the main stem	320	224	416
explicit parameter	PHOTOcoeff	unitless	strength of photoperiod response	50	35	65
explicit parameter	PHOTOcrit	h	critical photoperiod	13.50	7.00	17.55
explicit parameter	VERNreq	d	days to vernalisation saturation	49.00	34.30	63.70
implicit parameter	Ti1	unitless	intercept of the linear function for threshold induction temperature estimate	3.72	2.61	4.84
implicit parameter	Ti2	unitless	slope of the linear function for threshold induction temperature estimate	0.4011	0.2808	0.5214
implicit parameter	GDD1	unitless	coefficient for the estimate of minimum degree days to VRT requirement saturation	0.950	0.665	1.000
implicit parameter	GDD2	unitless	coefficient for the estimate of minimum degree days to VRT requirement saturation	340	238	442
implicit parameter	GDD3	°C ⁻¹	coefficient for the estimate of minimum degree days to VRT requirement saturation	2.0	1.4	2.6
explicit parameter	VERN_Tmin	°C	minimum temperature for vernalisation	-1.3	-3.3	0.7
explicit parameter	VERN_Topt	°C	optimum temperature for vernalisation	10	8	11

Category	Label	Unit	Definition Default		Lower limit	Upper limit
explicit parameter	VERN_Tmax	°C	maximum 12 temperature for vernalisation		11	14
implicit parameter	VRTfct1	unitless	VRT factor estimate 80 coefficient		56	104
implicit parameter	VRTfct2	unitless	VRT factor estimate coefficient	0.90	0.63	1.17
explicit parameter	RESPdays	d	minimum days of 10 snow cover duration to cause respiration		7	13
			stress			
explicit parameter	RESP_Tmin	°C	minimum value of the last 5- or 10-day average temperature for respiration stress	-1	-3	1
explicit parameter	RESP_Tmax	°C	maximum value of the last 5- or 10-day average temperature for respiration stress	1.5	1.0	3.5
explicit parameter	RESP_Tsd	°C	standard deviation of the last 5- or-10 days average temperature for respiration stress	0.750	0.525	0.975
implicit parameter	RESP1	unitless	coefficient for the estimate of respiration stress	0.540	0.378	0.702
implicit parameter	RESP2	unitless	coefficient for the estimate of respiration stress	0.840	0.588	1.092
implicit parameter	RESP3	unitless	coefficient for the estimate of respiration stress	0.0510	0.0357	0.0663
implicit parameter	RESP4	unitless	coefficient for the estimate of respiration stress	2.0	1.4	2.6
implicit parameter	RESP5	d ⁻¹	coefficient for the estimate of respiration stress	1.850	1.295	2.405
implicit parameter	DEHARD1	°C ² d ⁻¹	dehardening coefficient	5.050	3.535	6.565
implicit parameter	DEHARD2	°C	dehardening coefficient	4.350	3.045	5.655
implicit parameter	DEHARD3	unitless	dehardening coefficient	0.280	0.196	0.364
implicit parameter	LOWT1	°C ⁻¹	coefficient for the estimate of low temperature stress	-0.6540 - 0.8502		-0.4578
implicit parameter	LOWT2	d	coefficient for the estimate of low temperature stress	3.740	2.618	4.862

Category	Label	Unit	Definition	Default	Lower limit	Upper limit
implicit parameter	PHOTO1	unitless	photoperiod 3.50		2.45	4.55
			requirement estimate			
			coefficient			
implicit parameter	ΡΗΟΤΟ2	h⁻¹	photoperiod	0.5040	0.3528	0.6552
			requirement estimate			
			coefficient			
implicit parameter	РНОТОЗ	°C-1	photoperiod	0.3210	0.2247	0.4173
			requirement estimate			
			coefficient			
implicit parameter	PHOTO4	°C	photoperiod	13.2420	9.2694	17.2146
			requirement estimate			
			coefficient			
implicit parameter	PHOTO5	unitless	photoperiod	3.250	2.275	4.225
			requirement estimate			
			coefficient			
implicit parameter	HARDrate	°C-1 d⁻¹	hardening rate	0.0140	0.0098	0.0182

Туре	Symbol	Unit of measure	Definition
State variable	LT ₅₀	°C	lethal temperature 50%
Rate variable	ΔLT_{50}	°C d ⁻¹	LT ₅₀ rate
Rate variable	$\Delta LT_{50Hflow}$	°C d ⁻¹	actual hardening rate
Rate variable	$\Delta LT_{50D flow}$	°C d ⁻¹	actual de-hardening rate
Rate variable	$\Delta LT_{50R flow}$	°C d ⁻¹	loss of frost tolerance due to
			respiration stress rate
Rate variable	$\Delta LT_{50Sflow}$	°C d ⁻¹	loss of frost tolerance due to low
			temperature stress rate
Auxiliary variable	Ti	°C	threshold induction temperature
Auxiliary variable	$\Delta LT_{50H rate}$	°C d ⁻¹	hardening rate
Auxiliary variable	$\Delta LT_{50D rate}$	°C d ⁻¹	de-hardening rate
Auxiliary variable	LT _{50adj}	°C	damage-adjusted LT ₅₀
State variable	acclAmt	°C	accumulated acclimation
State variable	dehardAmt	°C	accumulated amount of
			dehardening due to $T_c > T_i$
State variable	respProgress	°C	accumulated amount of
			dehardening due to respiration
			stress
State variable	dehardAmtStress	°C	accumulated amount of
			dehardening due to stresses
Auxiliary variable	VRTprogress	unitless	progress to
			vegetative/reproductive
			transition
Auxiliary variable	VRTfactor	unitless	vegetative/reproductive
			transition factor
Auxiliary variable	photoProg	unitless	progress to photoperiod
	_		requirement saturation
State variable	vernDays	d	vernalization days
State variable	vernProg	d	progress to vernalisation
			requirement saturation
Auxiliary variable	vernSaturation	unitless	progress to vernalisation
Determinately	17	ac 1-1	requirement saturation
Rate variable	LI _{50 min flow}		rate of the state variable L1 _{50 min}
State variable	LI _{50 min}	<u>°C</u>	minimum value reached by L1 ₅₀
Auxiliary variable	DDReqCurrentTemp	unitless	growing degree days
	CI		requirement
Rate variable	<i>mfIn_{flow}</i>	unitless	rate of the state variable
			mjinFraction
State variable	mfinFraction	unitiess	progress towards minimum final
Deteveriable	veraDate		
	vernkale	unitiess	
Auxiliary variable	photoFactor	unitiess	progress to photoperiod
Pato variable	nhoto	unitlass	rate of the state variable
	photoflow	unitiess	nhotoReaFraction
State variable	nhotoReaFraction	unitlass	progress to photoporiod
State validule	μποτοπειγγιατίτοπ	unitiess	progress to photoperiou
			requirement saturation

Table 2. Variables of the model by Byrns et al. (2020).

Site	Coarse materials (%)	Clay (%)	Sand (%)	Silt (%)	Soil organic carbon concentration (g kg ⁻¹)
Sant'Angelo Lodigiano (Italy)	0	18	49	33	25
Merzenich (Germany)	6	24	13	63	10
Karklupėnai (Lithuania)	6	13	24	63	165

Table 3. Soil characteristics (0-20 cm) in the three sites.

Table 4. Statistics of the simulated soil temperature for the time frame between sowing date and the end April. The average soil temperature of the coldest or warmest month was obtained by averaging the daily soil temperatures of the considered month for the 20 years of simulation.

Site	Sowing date	Average soil temperature of the coldest month (°C)	Minimum soil temperature of the coldest month (°C)	Average soil temperature of the warmest month (°C)
Sant'Angelo	SD1	3.01 (January)	-5.70 (February)	22.47 (September)
Lodigiano (Italy)	SD2	3.37 (January)	-5.64 (February)	15.67 (October)
	SD3	3.60 (January)	-5.46 (February)	13.74 (April)
Merzenich	SD1	3.19 (January)	-11.61 (December)	17.94 (September)
(Germany)	SD2	3.43 (January)	-11.77 (December)	12.84 (October)
	SD3	3.64 (January)	-11.86 (December)	10.85 (April)
Karklupėnai	SD1	-0.99 (January)	-15.73 (January)	15.88 (September)
(Lithuania)	SD2	-0.78 (January)	-15.81 (January)	9.97 (April)
	SD3	-0.41 (January)	-10.99 (January)	10.77 (April)

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