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# The impacts of pollution sources and temperature on the light absorption of HULIS were revealed by UHPLC-HRMS/MS at the molecular structure level

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**ABSTRACT.** Atmospheric humic-like substances (HULIS), a key component of brown carbon (BrC), significantly promote the light absorption of aerosols. However, their linkages to pollution sources and ambient temperature in cold environments remain unresolved. Here, we analyze wintertime urban aerosol samples in Changchun, Northeast China, using ultrahigh performance liquid chromatography coupled with high-resolution tandem mass spectrometry (UHPLC-HRMS/MS). HULIS show a high light absorption efficiency ($MAE_{365}$ = 1.81 ± 0.24 $m^2$ $gC^{-1}$) and high mass concentration (2.97 ± 1.54 $\mu$gC $m^{-3}$), exceeding values reported from other global regions. Through UHPLC-HRMS/MS characterization, we identify 264 compounds at the molecular structure level, accounting for 38.2 - 78.1% of the total HULIS mass. Compositional analysis demonstrates biomass burning and coal combustion are the main BrC sources during haze events. We screen out 39 strong BrC chromophores, mainly nitrophenols, that contribute 28.9 ± 10.4% of the total light absorbance at 365 nm. Low ambient temperatures potentially enhance the accumulation of these strong BrC chromophores in the aerosol particles by suppressing photobleaching processes and altering thermodynamic reaction equilibria. These findings emphasize the potential of BrC to exert a more significant and persistent environmental effect in the cold region atmosphere.

**KEYWORDS:** *Humic-Like Substances, Non-targeted screening, Northeast China, Light absorption efficiency, Brown carbon chromophores*.

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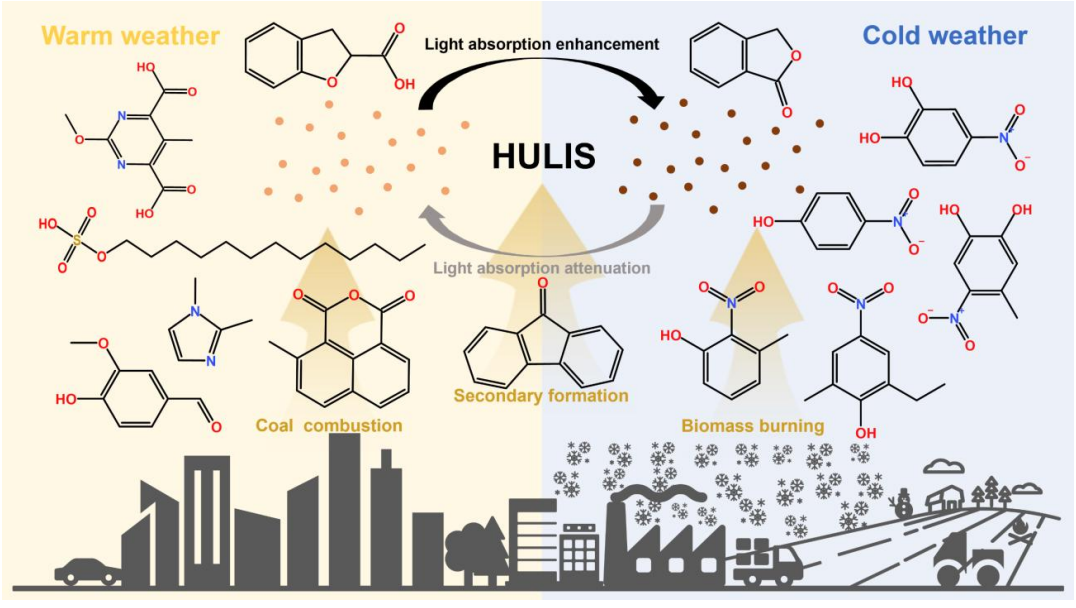
## Graphical Abstract



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## 1. Introduction

Atmospheric humic-like substances (HULIS) are important components in light-absorbing aerosols (Hoffer et al., 2006; Zou et al., 2023), therefore affecting global radiative forcing and atmospheric chemical processes (Chung et al., 2012; Huang et al., 2020; Laskin et al., 2015). They are identified as a highly complex aggregate of polar organic compounds composed of aromatic, aliphatic, and alicyclic structures with functional groups such as hydroxy, carbonyl, carboxyl, nitrooxy, and sulfooxy (Song et al., 2018; Wang et al., 2019; Zou et al., 2023). Previous studies have revealed that the molecular composition of HULIS determine their physicochemical properties, further impacting their climatic and environmental effects, such as cloud condensation nuclei activation, human health, and global radiation (Bao et al., 2023; Cappiello et al., 2003; Chen et al., 2021; Dou et al., 2015; Hems and Abbatt, 2018; Krivácsy et al., 2000).

As reactive components in the atmosphere, HULIS exhibit pronounced chemical activity through their oxygenated functional groups, particularly prone to the oxidation by reactive oxygen radicals and gaseous oxidants (Huo et al., 2021; Qiu et al., 2024). Both laboratory simulations and filed observations have demonstrated that these aging processes significantly alter the light-absorption features and environmental behavior of HULIS (Hems and Abbatt, 2018; Qiu et al., 2024; Wang et al., 2022, 2019). Although extensive investigations have characterized the elemental composition of HULIS (Lin et al., 2012; Song et al., 2018, 2022a; Wang et al., 2017a, 2019; Zou et al., 2023), critical knowledge gaps persist regarding the molecular structures determining their light absorption. This limits comprehension understanding of the atmospheric evolution process and radiative effect of HULIS.

Following emissions into the atmosphere, HULIS undergo vertical transport accompanied by sharp ambient temperature declines (Chen et al., 2021; Slade et al., 2017). This indicates that the atmospheric evolution of HULIS is under low-temperature conditions during their majority lifetime. How low temperature impact the atmospheric evolution process of HULIS remains uncertain yet. The decrease in temperature could potentially alter the physicochemical properties of HULIS, influencing their volatility (Cao et al., 2018; Schervish and Donahue, 2020), reactivity (Liu et al., 2023; Slade et al., 2017), and partitioning between the gas and particulate phase (Arp et al., 2008; Tao and Murphy, 2021). Consequently, their light absorption and atmospheric lifetime might be profoundly affected (Gregson et al., 2023; Roelofs, 2013). Therefore, it is imperative to further explore the low-temperature behavior of HULIS through field observations in cold environments.

This study collected atmospheric $PM_{2.5}$ samples in Changchun, which experiences low temperatures in wintertime, and HULIS were subsequently extracted. By employing a combination of non-targeted analysis and ultrahigh performance liquid chromatography coupled with high-resolution tandem mass spectrometry (UHPLC-HRMS/MS), we aimed to identify the molecular structures in HULIS collected from urban aerosols during wintertime. This approach provided us new insights into the potential sources and temperature effects on the light-absorption properties of HULIS.

## 2. Experimental Section

**2.1 Aerosol sampling and HULIS extraction.** We conducted a field campaign on the campus of Jilin University in Changchun, Northeast China (125.29° E, 43.83° N) from January 1$^{st}$ to 30$^{th}$, 2023. During this period, a high-volume particulate sampler (Tianhong Intelligent Instrument Plant, Wuhan, China, 1.05 $m^3$ $min^{-1}$) collected 24 h $PM_{2.5}$

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samples on a pre-baked quartz filter. **Figure S5** presented the meteorological data (http://www.wunderground.com/) and air pollutants data (http://air.cnemc.cn:18007/), including relative humidity, temperature, concentrations of CO, $SO_2$, $O_3$, $NO_2$, $PM_{2.5}$, and $PM_{10}$ during the sampling campaign.

The preparation process of HULIS sample was identical with previous studies (Limbeck et al., 2005; Yuan et al., 2021; Zou et al., 2020), and can be briefly described as the following steps: sampled filter was firstly extracted with ultrapure water (> 18.2 MΩ) in an ultrasonic bath for 40 mins. After that, the water extracts were filtered through 0.22 $\mu$m PES syringe filters, then acidified to pH=2 by HCl solution (0.1 M) and loaded on the pre-acidification solid phase extraction (SPE) cartridges (Supelclean ENVI-18, 500 mg, 3 mL). The majority of inorganic ions, low molecular-weight organic acids, and sugars were eluted out with ultrapure water while the fractions retained in the SPE cartridge were eluted with methanol (Baduel et al., 2009). Finally, a portion of the elution was measured by UHPLC-HRMS/MS and the rest was dried under a gentle stream of $N_2$, then redissolved in ultrapure water for total organic carbon and light absorption analysis.

**2.2 Molecular composition analysis of HULIS.** An ultrahigh performance liquid chromatography system (UHPLC, Dionex Ultimate 3000, Thermo Fisher Scientific, San Jose, CA, U.S.A.) coupled with an Orbitrap Fusion Tribrid mass spectrometer (Thermo Fisher Scientific, San Jose, CA, U.S.A.) was used to detect the molecular composition of HULIS. To detect as many HULIS species as possible and achieve quantification, we optimized the detection method to decrease the method detection limit and applied a semi-quantitative strategy to quantify the identified compounds (detailed in **Text S1**).

The optimized chromatographic conditions were as follows: Accucore C18 2.6 $\mu$m particle size (100 × 2.1 mm, Thermo Scientific) with the gradient elution started from 80% of mobile phase A (0.05% acetic acid) with a 0.2 mL $min^{-1}$ flow rate for 2 min, then changed to 100% of mobile phase B (methanol with 0.05% acetic acid) in 15 min and maintained constant for 2 min, decreased to 20% of mobile phase B within 1 min and finally held for 3 min for re-equilibration. The mass spectra (*m/z* 60-600) with a resolving power of 120,000 (*m/z* 200) were obtained by using heated-electrospray ionization (H-ESI). The optimized mass spectrometric parameters were as follows: 3.5 kV spray voltage for positive ions and 3.25 kV spray voltage for negative ions, 35 psi sheath gas (nitrogen), and 10 psi auxiliary gas (nitrogen), 320 °C ion transfer tube temperature, 125 °C vaporizer temperature. The data acquisition used data dependent mode and the master scans interval time was set as 1.0 second for the full scan experiments (detailed in **Table S4**).

**2.3 Light absorption analysis of HULIS and other analysis.** A total of the HULIS extract was first diluted to 3 mL with ultrapure water and then measured by a UV-Vis spectrophotometer (UV-1900, Shimadzu, Kyoto, Japan) at 200-700 nm with an interval wavelength of 1 nm. To assess the optical properties of HULIS samples, the mass absorption efficiency ($MAE_\lambda$ , $m^2$ $gC^{-1}$) was calculated according to the following formula.

$$Abs_\lambda = (A_\lambda - A_{700}) \frac{V_l}{V_a \times l} \ln 10 \qquad (1)$$

$$MAE_\lambda = \frac{Abs_\lambda}{C} \qquad (2)$$

where $Abs_\lambda$ represents the light absorption coefficient of the HULIS extract at a wavelength of $\lambda$ nm ($Mm^{-1}$), $A_\lambda$ is the recorded absorbance value of the HULIS extract by the UV-Vis spectrophotometer, $V_l$ is the total solution volume

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of HULIS extract (mL), $V_a$ is the air sampling volume corresponding to the volume of HULIS extract ($m^3$), $l$ represents the optical path length (0.01 m), $C$ is the mass concentration of HULIS carbon (HULIS-C) ($\mu$gC $m^{-3}$).

The contents of elemental carbon (EC) and organic carbon (OC) in quartz fiber filters were determined by a Thermo-Optical Transmission (TOT) method on a Sunset Lab EC/OC analyzer. The concentrations of HULIS-C were analyzed by a total organic carbon analysis (TOC-L, Shimadzu, Kyoto, Japan). The water-soluble inorganic ions in $PM_{2.5}$ collected on Teflon filter were detected by ion chromatography (IC, Shimadzu, Kyoto, Japan). The aerosol liquid water content (ALWC) and pH were calculated using the ISORROPIA II thermodynamic model based on meteorological data and mass concentrations of water-soluble inorganic ions (Fountoukis and Nenes, 2007; Nenes et al., 1998; Wu et al., 2018).

## 3. Results and Discussion

**3.1 Molecular composition and light absorption of HULIS. Figure 1A** illustrated the temporal variations of HULIS-C, OC, and EC mass concentrations over the entire sampling period during which temperatures ranged from 2.9 to -25.3 ℃ and solar radiation ranged from 24.3 to 57.8 W $m^{-2}$ (**Figure 1B**). The average concentrations of OC and EC were 11.7 ± 5.74 and 2.06 ± 0.92 $\mu$g $m^{-3}$, respectively. The average HULIS-C concentration amounted to 2.97 ± 1.54 $\mu$g $m^{-3}$, constituting 25.1% of the total OC. The observed HULIS-C concentration exceeded levels documented in winter of Europe (0.68 – 1.47 $\mu$g $m^{-3}$) (Emmenegger et al., 2007; Voliotis et al., 2017), South America (0.68 – 1.47 $\mu$g $m^{-3}$) (Serafeim et al., 2023), Chinese other regions (1.96 – 2.38 $\mu$g $m^{-3}$) (Lu et al., 2019; Ma et al., 2019; Zou et al., 2023), indicating the abundance of HULIS in Changchun.

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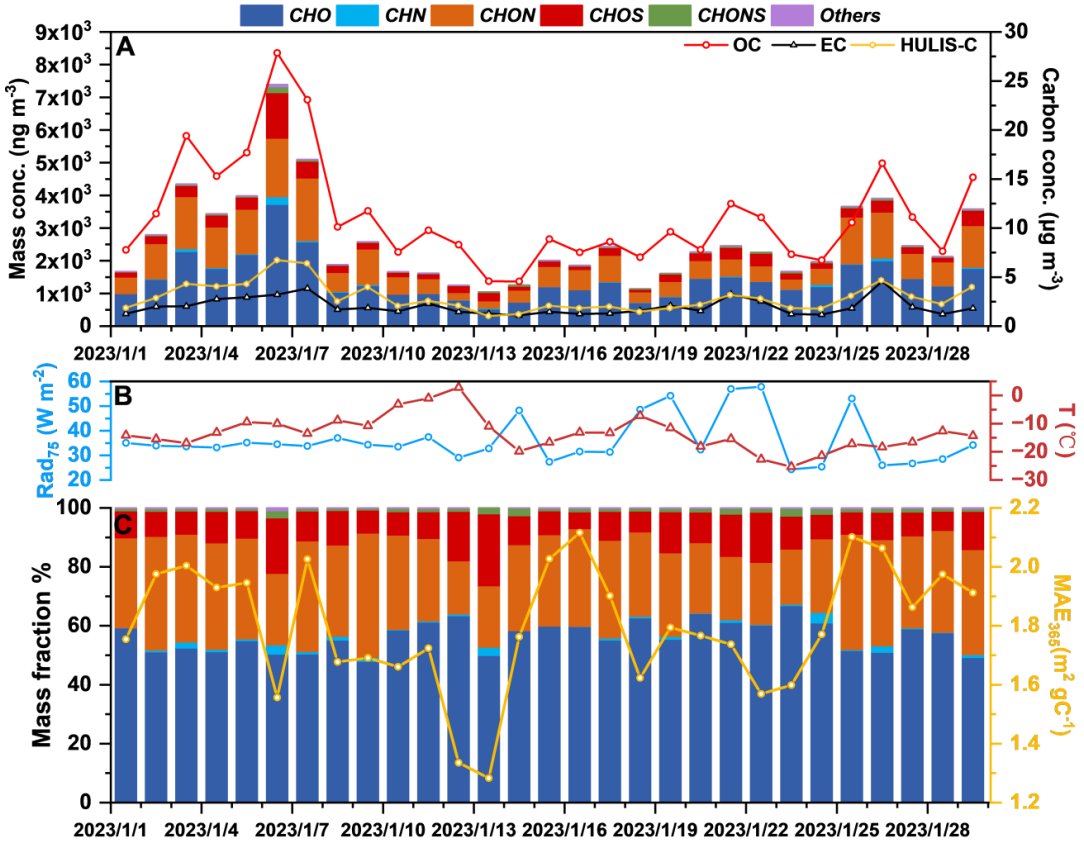


**Figure 1.** Temporal variations in the mass concentrations of six compound categories, organic carbon, and elemental carbon (A); the 75$^{th}$ percentile of solar radiation ($Rad_{75}$) and ambient temperature (B); the mass fraction of six compound categories as well as $MAE_{365}$ (C).

Molecular composition analysis of HULIS identified 264 compounds at the molecular structure level. **Table S6** in the supplement listed the details of these HULIS compounds and their corresponding confidence levels. The identified compounds were grouped into six compound categories based on their elemental types, including CHO, CHN, CHON, CHOS, CHONS, and other species. CHONS category refers to compounds that contain carbon, hydrogen, oxygen, nitrogen, and sulfur elements. **Figure 1A and 1C** showed variations of six compound categories. The total mass concentration of these compounds ranged from 1.05 to 7.39 $\mu$g m$^{-3}$ (**Figure 1A**), explaining 38.2% - 78.1% of the total HULIS mass (converted by multiplying [HULIS-C] by 1.6, (Friman et al., 2023)). The remaining unidentified compounds in HULIS mainly include low-polarity phenols, ketones, and aldehydes, whose polarity is too low to be ionized in the ESI mode (Huang et al., 2025; Huo et al., 2021; Song et al., 2022b, 2024).

**Figure 1C** also displayed that MAE at 365 nm ($MAE_{365}$) of HULIS samples. The $MAE_{365}$ ranged from 1.28 to 2.12 m$^2$ gC$^{-1}$ (1.81 ± 0.24 m$^2$ gC$^{-1}$ in average), which was higher than those in Beijing (1.79 ± 0.24 m$^2$ gC$^{-1}$) (Cheng et al., 2011), Xi'an (1.65 ± 0.36 m$^2$ gC$^{-1}$) (Huang et al., 2018), Guangzhou (1.1 ± 0.27 m$^2$ gC$^{-1}$) (Zou et al., 2023), and Hong Kong (0.97 ± 0.40 m$^2$ gC$^{-1}$) (Ma et al., 2019) during wintertime. This indicated that HULIS in Changchun have

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higher light absorption efficiency compared to other regions in China. Moreover, the strongly positive correlation (**Figure S6**) between $MAE_{365}$ with CHON category (Pearson's R = 0.77, *p*-value < 0.01) and aromatic fraction (Pearson's R = 0.86, *p*-value < 0.01) suggested that the high light absorption efficiency of HULIS may be related to aromatic CHON compounds.

**3.2 Potential sources of HULIS based on molecular analysis.** To analyze the cause of the high concentrations and light absorption efficiency of HULIS in this study, we selected two typical haze events with significant differences in $MAE_{365}$ (Event I: $PM_{2.5}$ = 159.6 ± 53.8 $\mu$g $m^{-3}$, $MAE_{365,HULIS}$ = 1.56 $m^2$ $gC^{-1}$; Even II: $PM_{2.5}$ = 83.7 ± 36.4 $\mu$g $m^{-3}$, $MAE_{365,HULIS}$ = 2.06 $m^2$ $gC^{-1}$) for potential sources comparison from the perspective of molecular composition. **Figure 2** exhibited the reconstructed MS spectra, the number, and concentration fraction of HULIS in both positive and negative modes.

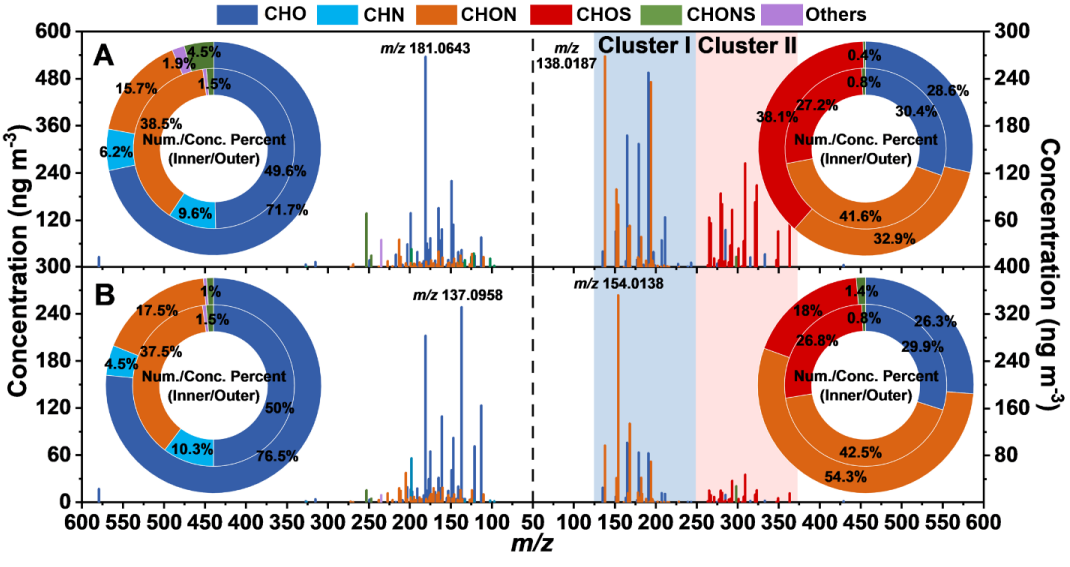


**Figure 2.** Reconstructed mass spectra (positive ions on the left, while negative ions on the right) for the identified HULIS samples during Event I (A) and Event II (B). The highest peaks were marked by their *m/z*. The inner and outer ring pie charts were the fraction of mass concentration and numbers in the different categories of the identified compounds, respectively.

In the positive mode, Event I and II had similar elemental composition, both dominated by CHO compounds, followed by CHON, CHN, and others. The most abundant species in Event I and II were 9-fluorenone (*m/z* 181.0643) and 2-[(1E)-1-Buten-1-yl]-5-methylfuran (*m/z* 137.0958), respectively. The former originates from diverse combustion sources such as biomass burning, coal combustion, and vehicle emission (Alves et al., 2016; Huo et al., 2021; Ma et al., 2023; Souza et al., 2014; Xu et al., 2024; Zhao et al., 2020), whereas the latter is believed to stem specifically from biomass burning (Bhattu et al., 2019; Hatch et al., 2015). Further evidence for the significant role of biomass burning and coal combustion in both events was provided by the high concentrations of biomass burning tracers (i.e. vanillin, syringaldehyde, acetosyringone in **Table S6**, and $K^+$ in **Table S5**) and $SO_2$ (**Table S5**) (Chen et al., 2017; Dutton et al., 2009; He et al., 2010; Liang et al., 2021). Biomass burning and coal combustion have been confirmed in our previous study to be the main sources of air pollution in Changchun winter (Dong et al., 2023).

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In the negative mode, two distinct compound clusters were observed within the *m/z* range of 125 – 250 (refer to Cluster I) and 250 – 375 (refer to Cluster II), as marked in the right part of **Figure 2**. Cluster I comprised a significant proportion of strong BrC species, such as nitrophenols (including 4-nitrophenol, 3-nitrocatechol, 4-nitro-1-naphthol, and etc., **Table S6**), mainly originating from primary emissions like biomass burning and coal combustion (Huang et al., 2023; Jiang et al., 2023; Lin et al., 2017; Wang et al., 2020). Notably, Cluster I fraction was more prominent in Event II than Event I, explaining the higher $MAE_{365}$ in the former.

All of CHOS compounds were characterized by ion fragment *m/z* 96.9595 in the MS/MS spectra and were therefore identified as organosulfates (OSs). Considering the OSs are typically formed by atmospheric aqueous reaction (Brüggemann et al., 2017; Pratt et al., 2013; Wach et al., 2020), we therefore proposed that Cluster II which was mainly composed of OSs originated from secondary formation. The higher abundance of Cluster II in Event I suggested that the secondary formation of HULIS was more intense during this event compared to Event II. The elevated concentrations of secondary inorganic ions (including $NH_4^+$, $NO_3^-$, and $SO_4^{2-}$, 11.35 – 27.66 vs 4.49 – 7.61 $\mu g\ m^{-3}$) and relative humidity (83.1 ± 4.6% vs 61.9 ± 14.0%) in Event I than those in Event II were observed, as detailed in **Table S5**. As a result, the higher ALWC (95.9 vs 23.9 $\mu g\ m^{-3}$) and lower pH value (4.20 vs 4.82) in Event I in contrast to Event II facilitated the formation of OSs. Since the OSs studied here were primarily aliphatic sulfates (summarized as the molecular formulas of $C_nH_{2n+2}O_{4-6}S$ and $C_nH_{2n}O_{4-6}S$, where $10 \leq n \leq 18$), which belong to non-light-absorbing organic matter, this may cause the lower $MAE_{365}$ value in Event I.

**3.3 Effect of ambient temperature on the BrC chromophores of HULIS.** As above-mentioned, the temperature was down to -25°C. Such low temperature may affect the evolution of HULIS in the atmosphere. In total, 39 compounds were screened as strong BrC chromophores to analyze the effect of temperature on the BrC chromophores according to a partial least squares regression (PLS) model (detailed in **Text S4**). These compounds belong to nitrophenols or nitrophenol derivatives, which are marked in **Table S6**. The 39 strong BrC chromophores accounted for 8.67 ± 3.68% of the total mass and contributed 28.9 ± 10.4% of the light absorbance (**Figure S7**), with an average $MAE_{365}$ of 7.40 ± 1.80 $m^2\ gC^{-1}$, indicating their importance in the light absorption of HULIS.

**Figure 3** presented that the mass fraction of screened 39 BrC chromophores, along with the $MAE_{365}$ value of HULIS, across different temperature ranges. Both mass fraction and $MAE_{365}$ increased with decreasing ambient temperature, indicating that low temperature may facilitate the accumulation of strong BrC species in the particle phase. We proposed two possible explanations: firstly, the low temperature may lead to a non-liquid phase state of ambient particles, potentially introducing kinetic limitation on the diffusion of reactive species from gas phase into particle bulk (Li and Shiraiwa, 2019). We utilized an established parameterization scheme (**Text S3**) to calculate the glass transition temperature (Tg) of HULIS based on their molecular composition (Li et al., 2020). The results showed that the decrease in ambient temperature (T) enhanced the Tg/T ratio, driving the phase transition of particles from liquid state (Tg/T = 0.76) to semi-solid state (Tg/T > 0.79). This may lead to the diffusion coefficients reduction of reactive species (Arangio et al., 2015; Gatzsche et al., 2017; Mikhailov et al., 2009; Shiraiwa et al., 2011; Virtanen et al., 2010), thereby slowing the degradation rate of BrC via hydroxyl radical oxidation or triplet excitation pathways in the atmosphere (Schnitzler et al., 2022; Schnitzler and Abbatt, 2018). These findings suggest that the non-liquid particle phase state, in conjunction with the weak solar radiation during Changchun's winter (refer to **Figure 1B**), results in a less pronounced photochemical aging of BrC, thereby diminishing its photobleaching.

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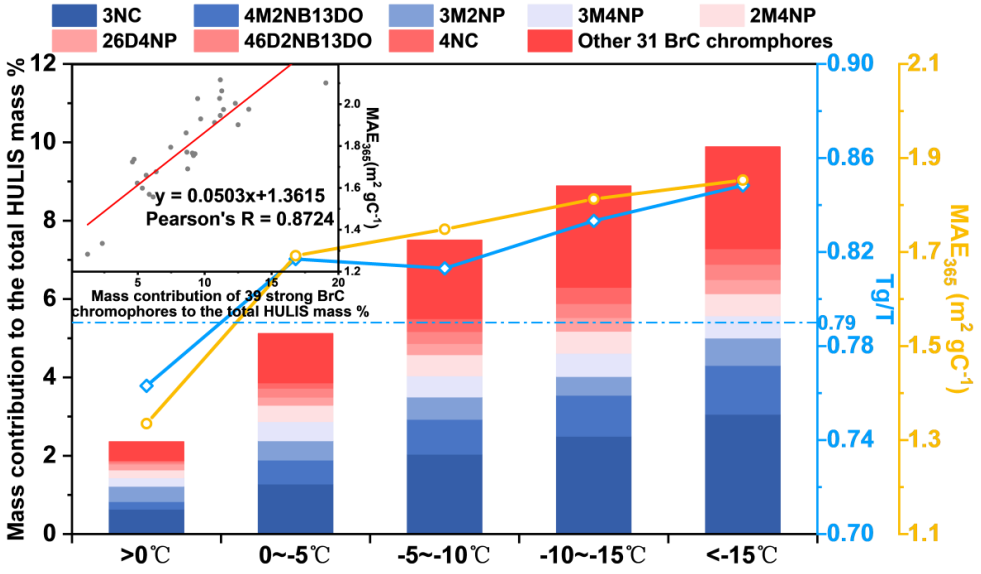


**Figure 3.** The mass contribution of 39 strong BrC chromophores, $MAE_{365}$ values, ratios of glass transition temperature (Tg) to temperature (T) at different temperatures, and linear relationships between mass contribution of strong BrC and $MAE_{365}$ of HULIS. The blue dotted line represented the threshold of Tg/T between liquid and semi-solid state (Shiraiwa et al., 2017), and the abbreviations of 3NC, 4M2NB13DO, 3M2NP, 3M4NP, 2M4NP, 26D4NP, 46D2NB13DO, and 4NC represents 3-nitrocatechol, 4-methyl-2-nitrobenzene-1,3-diol, 3-methyl-2-nitrophenol, 3-methyl-4-nitrophenol, 2-methyl-4-nitrophenol, 2,6-dimethyl-4-nitrophenol, 4,6-dimethyl-2-nitrobenzene-1,3-diol, and 4-nitrocatechol, respectively.

Secondly, the formation of BrC chromophores was also important for the $MAE_{365}$ enhancement of HULIS. On the one hand, the secondary formation of nitrophenols has been conclusively attributed to reaction of phenols with $NO_x$ radicals (Bolzacchini et al., 2001; Finewax et al., 2018; Kroflič et al., 2021; Mayorga et al., 2021), a process that has been characterized as exothermic (Bolzacchini et al., 2001; Domingo et al., 2021). On the other hand, we have demonstrated that further atmospheric oxidation of nitrophenols proceeds via a ring-opening mechanism of benzene moiety (Qiu et al., 2024), which constitutes an endothermic reaction (Cao et al., 2021; Hems and Abbatt, 2018; Wang et al., 2017b). From a thermodynamic perspective, low temperature not only promotes exothermic chemical reactions but also hinders endothermic processes, thereby accumulating the strong BrC chromophores such as nitrophenols in HULIS. As such, we infer that ambient temperature play a critical role in promoting the transformation and light absorption of BrC chromophores, particularly in cold or/and high-altitude regions.

**3.4 Conclusions.** In this work, we explored the linkage between the light absorption and molecular structure of atmospheric HULIS in Changchun winter based on UHPLC-HRMS/MS. In different haze events, the molecular structure of HULIS varied due to different sources, which lead to differences in their light absorption efficiency. Biomass burning and coal combustion were important inducers of the high $MAE_{365}$ value of HULIS. It was the fact that biomass burning and coal combustion emitted a large fraction of BrC chromophores such as nitrophenols, while aliphatic organosulfates produced by secondary formation lead to the reduction in the light absorption efficiency of HULIS.

In addition, our study screened out 39 strong BrC chromophores belonging to nitrophenols by PLS model. These

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species accounted for 8.67 ± 3.68% of the total HULIS mass while contributed nearly 30% of the light absorbance at 365 nm. We found that low temperature can promote the accumulation of strong BrC chromophores through slowing down the photobleaching reaction and changing the thermodynamic reaction equilibrium, thereby improving the light absorption capability of HULIS. This phenomenon has not been studied before, and further laboratory and field studies are urgently needed to verify the effect of temperature on the light absorption properties of BrC.

Our research has found that in the cold regions of northern China, on one hand, primary emissions from biomass burning and coal combustion are relatively strong, and on the other hand, low temperatures reduce the photobleaching of brown carbon (BrC). This implies that BrC in cold regions may have a longer lifetime and stronger light-absorbing properties in the atmosphere, thus playing a more significant role in the direct radiative forcing of carbonaceous aerosols.

**Supplementary Information.** Optimization details of LC-MS method, calculation procedure of relevant index, screening details of PLS model, and analysis results of pollutant data and meteorological data (PDF).

The information about atmospheric mass concentration, molecular information, and strong brown carbon chromophores of identified compounds in HULIS samples of Changchun during wintertime (Table S6).

**Author Contributions.** T. Q. and Y. Q. designed this work. T. Q., X. W., and Y. G., collected the experimental samples. T. Q., Y. Q., Y. Y., R. S., Y. N., X. H., and X. M. collected and analyzed the experimental data. Z. W., D. L., D. D., and H. C. edited the manuscript. All authors have read and agreed to the published version of the manuscript.

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**Notes.** The authors declare no competing financial interest.

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