Evidence for amino acids in the gas of the IC 348 star cluster of the Perseus Molecular Cloud

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Abstract

Amino acids are building-blocks of proteins, basic constituents of all organisms and essential to life on Earth. They are present in carbonaceous chondrite meteorites and comets, but their origin is still unknown. Formation of amino acids in the interstellar medium is possible via specific gas-phase reactions in dark clouds, however sensitive radio searches at millimeter wavelengths have not revealed their existence yet. The mid-IR vibrational spectra of amino acids provides an alternative path for their identification. We present Spitzer spectroscopic observations in the star-forming region IC 348 of the Perseus Molecular Cloud showing evidence for mid-IR bands of $\text{H}_2\text{O}$, $\text{OH}$, $\text{H}_2\text{O}$, $\text{CO}_2$, $\text{C}_2\text{H}_2$, $\text{C}_6\text{H}_6$, $\text{C}_6\text{H}_5\text{N}$, $\text{PAHs}$, fullerenes C60 and C70 and emission lines consistent with the most intense laboratory bands of the three aromatic amino acids, tyrosine, phenylalanine and tryptophan and the aliphatic amino acids isoleucine and glycine. Estimates of column densities give values 10-100 times higher for isoleucine and glycine than for the aromatic amino acids as in some meteorites. The strongest bands of each amino acid are also found in the combined spectrum of >30 interstellar locations in diverse star-forming regions supporting the suggestion that amino acids are widely spread in interstellar space. Future mid-IR searches for proteinogenic amino acids in protostars, protoplanetary disks and in the interstellar medium will be key to establish an exogenous origin of meteoritic amino acids and to understand how the prebiotic conditions for life were set in the early Earth.

1. INTRODUCTION

Proteinogenic and non-proteinogenic amino acids have long been known to be present in meteorites, especially carbonaceous chondrites (Kvenholden et al. 1970, Cronin and Moore 1971, Cronin & Chang 1993, Ehrenfreud & Charnley 2000) and their abundances have been compiled in numerous studies (e.g. Commeyras et al. 2005, Martins & Sephton 2009, Martins 2011, Cobb & Pudritz 2014, Pizzarello & Shock 2017; Koga & Naraoka 2017). The presence of amino acids in meteorites and comets (Elsila et al. 2009) suggests that the young planet Earth was enriched with these molecules, basic constituents of proteins, essential for the emergence of life and the development of living organisms (Chyba et al 1990, Botha and Bada 2002, Muñoz Caro et al. 2002).

A few dozen molecules have been detected in the interstellar medium of star-forming regions (Cazaux et al. 2003, Jørgensen et al. 2012, Kahane et al. 2013, Favrè et al. 2018) and in
protoplanetary disks (Dutrey et al. 2014). These detections include aldehydes, acids, ketones, and sugars. The simplest organic acid, formic acid (HCOOH), which contains the carboxyl group, one of the main functional groups of amino acids, has been detected in low-mass star-forming regions (Liu et al. 2002, Lefloch et al. 2017) and in a protoplanetary disk (Favrè et al. 2018). The reaction of protonated alcohols and HCOOH could lead to the production of glycine (and other amino acids) in the hot nuclei of molecular clouds (Ehrenfreund and Charnley 2001) and formation of amino acids in the interstellar medium is possible via specific gas-phase reactions in dark clouds (Ehrenfreund and Charnley 2000, Elsila et al. 2007, Martins and Sephton, 2009, Redondo et al. 2017). This suggests that amino acids could be present in star-forming regions, however, conclusive evidence has not been obtained yet. Very sensitive searches of amino acid bands conducted at millimeter wavelengths have focused on glycine, the most simple amino acid, and obtained upper limits to the column density (e.g. Ceccarelli et al. 2000) or tentative identifications of ro-vibrational bands (Kuan et al. 2003) which have not been finally confirmed (Snyder et al. 2005, Jones et al. 2007, Cunningham et al. 2007). Identifying the site and processes of amino acid formation in interstellar space remains a challenge of great importance in astrochemistry and astrobiology.

The mid-IR spectrum of amino acids offers an alternative way to explore their presence in the interstellar medium. Systematic laboratory work (e.g. Husain et al. 1984, Matei et al. 2005, Iglesias-Groth and Cataldo 2018) has led to the identification of a large number of relatively intense amino acid bands in the mid-IR. Between 15 and 20.8 µm these bands are mostly due to the carboxylate group COO⁻ rocking, bending and wagging modes; in the range 20.8 to 26.3 µm are mainly associated to the protonated amino group, i.e. NH₃⁺ vibration modes and, at longer wavelengths, between 26.3 and 37.0 µm the bands are mainly related to the C-Cα-N deformation modes (see e.g. Iglesias-Groth and Cataldo 2018). Weak hydrogen bond vibration bands have also been identified from 45.5 to 100.0 µm which may be useful to achieve a solid amino acid identification. The Spitzer Space Telescope has obtained a large, very valuable number of moderately high resolution spectra of star-forming regions in the spectral range 10-40 µm. Since band patterns significantly vary among amino acids in this spectral range, it appears particularly interesting for a potential identification of individual amino acid species (see e.g. Iglesias-Groth and Cataldo 2018).

The young stellar cluster IC 348 (age ≈ 2 Myr), located at the eastern end of the well known Perseus Molecular Cloud complex (Herbig 1998, Luhman et al. 2003, Bally et al 2008, Luhman et al. 2016) at a distance of 321 ±10 pc (Ortiz-León et al. 2018), is one of the nearest star-forming regions which has proved well suited to explore the presence of complex carbon-based molecules (see e.g. Iglesias-Groth 2019). The Perseus Molecular Cloud also provided one of the first unambiguous detections of the anomalous microwave emission (Watson et al. 2005), a radiation likely caused by electric dipole emission (Draine and Lazarian 1998) of a diffuse distribution of fast spinning carbonaceous molecules (polycyclic aromatic hydrocarbons, hydrogenated fullerenes, etc.). Organic molecules appear to be widely distributed in this nearby molecular cloud complex which prompted us to carry out an extensive search for amino acids.

Among all the 20 proteinogenic amino acids precise laboratory measurements of wavelengths, molar extinction coefficients and integrated molar absorptivities of mid-IR bands are available for the aromatic amino acids tryptophan, tyrosine and phenylalanine and for the aliphatic isoleucine and glycine (Iglesias-Groth and Cataldo 2021). In this work, we present results on the search in the
Perseus Molecular Complex for mid-IR bands (10-30 μm range) of these five amino acids.

2. OBSERVATIONS

We have used high spectral resolution (R=600) archive spectra obtained with the Infrared Spectrograph (IRS) onboard the Spitzer Space Telescope at various interstellar medium locations in the central region of IC 348 in the Perseus Molecular Cloud. All the selected observations were located within 10 arcmin of the most luminous star of the cluster, HD 281159, within the region marked in Fig. 1. Fully reduced spectra acquired with the Short-High (S-H, 9.8-19.5 μm) and Long-High (L-H, 19.5-36 μm) modules were taken from the Combined Atlas of Spitzer/IRS Sources (CASSIS; http://cassis.sirtf.com, Lebouteiller et al. 2015) which provides reduced and flux calibrated data. The spectra used from both modules of the IRS are identified with the corresponding AOR numbers in Table 1. Coordinates of the ISM pointings are also listed. The present search is focused on interstellar locations so we have selected pointings which in the original observing programmes, mainly focused on protoplanetary discs, where serving as background observations for nearby targets. Our interest in this first work is to explore the presence of amino acids in the ISM as disc continuum emission could limit the detectability of faint bands.

The S-H and L-H spectra are not available for the same interstellar locations, however we do not intend to carry out studies of individual locations, on the contrary, we will average the available spectra from different pointings in the core region of IC 348 from both IRS modules in order to gain as high signal to noise as possible to facilitate the search for weak bands. For comparison purposes, 32 spectra obtained with the same instrument at ISM locations in various star-forming regions, unrelated to the Perseus molecular complex, will be combined to generate a star-forming region “ISM reference” spectrum of similar high signal to noise to that of Perseus. The log of these observations can be found in Table 2 which lists AOR numbers and pointing coordinates for these Spitzer spectra. In this case, for each pointing there is full spectral coverage from 10 to 35 μm.

3. DATA

The CASSIS pipeline takes the Basic Calibrated Data (BCD) images, the associated uncertainty images and the bad pixel mask as starting products. The BCD images are produced by the Spitzer Science Center BCD pipeline. The High-resolution modules of the IRS are significantly affected by cosmic ray hits and spurious features, thus the CASSIS pipeline (Lebouteiller et al. 2015) pays special care to the exposure combination and image cleaning to remove any bad pixels in order to minimize these effects. This pipeline incorporates various improvements with respect to Lebouteiller et al. (2011) including several techniques for optimal extraction and a differential method that eliminates low-level rogue pixels.

As we are interested on observations of the general interstellar medium we will use the CASSIS full-aperture extraction for extended sources. In order to compute fluxes, this extraction method co-adds the pixels in the detector area corresponding to one wavelength value. Since the flux is integrated, the presence of bad pixels anywhere within this area is especially damaging and bad
pixels need to be picked out and replaced. The “cleaning” is carried out on the combined image of all exposures obtained for a given target. Bad pixel fluxes are substituted using neighbours whose flux is more reliable when exposures have been combined. The most problematic are the rogue pixels which have elevated dark current that change unpredictably with time. The CASSIS pipeline uses IRSCLEAN, an IDL tool for creating bad pixel masks from Spitzer IRS BCD (and pre-BCD) image data, and "clean" the masked pixels in a set of data to substitute bad pixels. As described by Leboutellier et al. (2015) this includes pixels with no values (NaNs), with a high bad pixel mask value (>256), bad pixels and rogue pixels flagged in the campaign mask, pixels with a large uncertainty and negative pixels.

The new pixel values calculated by the IRSCLEAN algorithm are mainly based on neighbouring pixels taking into account uncertainties and mask values propagated for each step. Subsequently, the full-aperture extraction is performed using the standard tool in SMART. An error on the flux is ultimately calculated using the quadratic sum of the pixel uncertainties in the detector area corresponding to each wavelength value. No background subtraction is applied for full-aperture extraction, however this is not expected to introduce any significant uncertainty on the measurement of wavelengths and fluxes of the relatively weak bands which we aim to detect.

As a check of the effectiveness of the bad pixel correction used in the full aperture extraction technique, we have compared the CASSIS full aperture and optimal differential extracted spectra for the well studied source RNO 90 (Pontoppidan et al 2010, Salyk et al. 2011). Figure 2 shows the spectra obtained by these two CASSIS extraction methods, small flux differences (typically less than 0.05 Jy) are seen in the continuum, probably due to the background correction applied by the optimal differential extraction. The corrections for bad pixels that both extraction techniques use do not seem to produce residual artifacts which could be confused with weak bands. If this happens, such artificial bands have peak-fluxes significantly below 0.05 Jy. As a further test we have measured fluxes for well known water bands in the full aperture spectrum of RNO 90 and compared them (see Table 3) with those reported by Blevins et al. (2016) from a completely independent reduction of the Spitzer RNO 90 spectrum. The comparison of band fluxes shows good agreement, with differences at the level of only a few percent of the measured fluxes.

The individual CASSIS full-aperture extraction of the S-H and L-H spectra available in the core of IC 348 showed very similar features and were subsequently averaged to produce a high signal to noise spectrum designated hereafter as “combined IC 348 ISM”. Similarly, we produced a star-forming region ISM “reference” spectrum by averaging the 32 spectra selected from ISM observations of other diverse star-forming regions. The average of the rms, $\sigma$, of fluxes recorded in various spectral regions (10.10-10.15 $\mu$m, 11.75-11.80 $\mu$m, 14.35-14.5 $\mu$m, 14.6-14.7 $\mu$m, 15.45-15.50 $\mu$m, 16.8-16.9 $\mu$m, 20.4-20.7 $\mu$m and 27.40-27.60 $\mu$m) was used to estimate the minimum flux level for a line detection in the combined IC 348 ISM spectrum which was adopted as the integrated flux of a line with peak flux of $2\sigma$ and FWHM=0.02 $\mu$m. This flux detection level resulted of order $1 \times 10^{-18}$ W m$^{-2}$. Line detectability at such low fluxes opens the possibility to identify weak transitions, enabling, in particular, a search for mid-IR transitions of the selected amino acids.
4. IDENTIFICATION OF ATOMIC AND MOLECULAR BANDS

As a first step towards the identification of amino acids in the gas of IC 348, it is important to search in the combined IC 348 ISM spectrum for bands of the most common atomic and molecular species reported in diverse interstellar environments. At mid-IR wavelengths there are fundamental vibrational transitions of many relevant molecules (e.g. H$_2$O, CH$_4$, C$_2$H$_2$, HCN, CO$_2$, etc) which could limit the detectability of amino acid bands. The HITRAN molecular spectroscopic database (see Gordon et al. 2020 for a most recent description) was used to explore the presence of molecular bands in the observed spectrum. The HITRAN Application Programming Interface (HAPI, Kochanov et al. 2016) was used for downloading and selecting spectroscopic transitions for each molecular species in the HITRAN online web server and for calculating the corresponding absorption coefficients and absorption spectra in the spectral range of interest (10-32 μm) based on the available line-by-line spectroscopic parameters. The computed spectra, as well as molecular mid-IR bands reported in the literature served as a reference for their identification in the spectrum of the interstellar gas in IC 348. In addition, mid-IR transitions of atomic species commonly detected in interstellar environments were considered in this search.

4.1 Molecular hydrogen emission, H$_2$

The IC 348 intermediate resolution IRS spectra allowed us to detect the rotational transition lines of molecular hydrogen, H$_2$ S(0) ($\nu$=0–0, J=2–0, 28.22 μm) and S(1) ($\nu$=0–0, J=3–1, 17.04 μm) lines. These lines are detected with > 10 σ amplitude sensitivity. At shorter wavelengths the upper level transition S(2), (S(2): $\nu$=0–0, J=4–2, 12.28 μm) is also detected. With far less confidence, the $\nu$=1–1 S(3) transition at 10.18 μm is also detected (peak flux 0.007 Jy relative to the local continuum). Fig. 3 shows the best linefits for all three detected (0-0) H$_2$ lines. These transitions are strong enough to provide good flux determinations. These fluxes were obtained with the IRAF SPLOT routine adopting appropriate local continuum for each of the lines. The quadrupole moment of the pure rotational 0–0 transitions of H$_2$ makes them direct tracers of the H$_2$ gas, these transitions are excited at higher temperatures than those typically prevalent in molecular cloud interiors and trace most of the warm molecular gas with temperatures between 100 and 1000 K. The populations of these levels are thermalized under most conditions and the S(0) and S(1) lines may provide a direct measure of the masses and temperature of the bulk of warm molecular gas at T = 50-200 K. The higher pure rotational lines, as well as the vibration – rotation lines at 2 μm, are probes of the photon or shock-heated gas (Draine and Bertoldi 1999).

4.1.1 H$_2$ column density and excitation temperature

The excitation diagram of H$_2$ rotational emission can be used to derive the level populations and excitation temperature of warm molecular hydrogen gas (e.g. Naslim et al. 2015). Using the measured line intensities $I_{obs}(u,l)$ of each transition, the H$_2$ column densities of the upper levels ($N_u$, given in Table 4) are derived from:

$$N_u = 4\pi \lambda I_{obs(u,l)} / (hc A_{ul}),$$  \hspace{1cm} (1)
where $A_{ul}$ is the Einstein coefficient for total radiative decay probability of the upper level. It is assumed that the lines are optically thin, the radiation is isotropic and the rotational levels of H$_2$ are thermalized. Under these assumptions, the level population follow the Boltzmann distribution law at a given temperature and the total H$_2$ column densities $N$(H$_2$tot) can be determined in the local thermodynamic equilibrium (LTE) condition using the formula:

$$N_u/g_u = [N(H_2)_{tot}/Z(T)] \exp \left(-\frac{E_u}{k_B T}\right), \quad (2)$$

where $Z(T) \approx 0.0247T/[1 - \exp(-6000K/T)]$ is the partition function (Herbst et al. 1996).

The statistical weight is $g_u = (2s + 1)(2j + 1)$, with spin number $s = 0$ for even $J$ (para-H$_2$) and $s = 1$ for odd $J$ (ortho-H$_2$) (Rosenthal, Bertoldi & Drapatz 2000). If the H$_2$ level populations are completely dominated by collisional excitation and de-excitation then the excitation temperature equals the kinetic temperature of the gas. Population diagrams of $\ln(N_u/g_u)$ versus $E_u/k_B T$ allow investigation of whether the levels are thermalized. If they are thermalized, the upper level column densities normalized with the statistical weight, $g_u$, in logarithmic scale as a function of the upper level energy, $E_u$, defines a straight line in the excitation diagram with a slope $1/T$ (Goldsmith & Langer 1999). If a range of gas temperatures occur along the line-of-sight, the points will lie on a curve with the lower energy levels lying on a steeper slope (lower temperature) than the higher energy levels (higher temperature). This is because the higher energy levels are preferentially populated at higher temperatures. In Fig. 4 we see that, most likely, this is the case. In reality the ISM is made of gas with a distribution of temperatures and a multiple-temperature fit is frequently needed for characterizing the excitation diagrams (see e.g. Naslim et al. 2015).

Since we only have three line detections (S(0), S(1), S(2)) the excited H$_2$ total column density, $N$(H$_2$) and excitation temperature, can be determined with modest precision. In a two-temperature fit, the cooler component results at temperature of 190 K (warm gas) and the second component of hotter gas appears at 385 K. Observations of higher J transitions (at shorter wavelengths) would be needed to obtain a precise determination of the excitation temperature of the hotter component. Extinction corrections to the measured fluxes are small and have not been applied, thus these excitation temperatures, which are valid for the purpose of this paper, should be taken only as indicative. If a single fit is applied to the three available measurements the average excitation temperature results $T = 258 \pm 10$ K and the associated total column density from expression 2 above is $N$(H$_2$) = $1.9 \times 10^{20}$ cm$^{-2}$ (uncertainty of 40%). Snow et al. (1994) measured a higher value of $N$(H$_2$) = $5.5 \times 10^{20}$ cm$^{-2}$ (and $N$(H I) = $2.6 \times 10^{21}$ cm$^{-2}$) from observations in the line of sight of the most luminous star in the central region of IC 348. These two independent measurements, which correspond to different regions of the ISM in IC 348, are rather similar. Applying a moderate extinction correction to our fluxes will make them even more consistent.

In summary, the strength of the H$_2$ bands indicate in the averaged spectrum of the IC 348 interstellar regions the presence of at least two warm gas components, of order 200 and 400 K, respectively. Most of the H$_2$ in the regions under study appear to be associated with the 200 K component.

4.1.2 HD
The heavier isotope, HD, has a small dipole moment (approx. $10^{-4}$ Debye) and its lowest J=1-0 line at 112 µm was already detected with ISO (Wright et al. 1999). The (0-0) R(5) J=6-5 line at 19.5 µm was observed by Bertoldi et al. (1999) in the Orion shock and in combination with H$_2$ data used to determine the D/H ratio in this star-forming region resulting in [D]/[H] = 1-2 x $10^{-5}$.

The IC 348 spectrum was searched for the presence of the series of pure rotational HD lines 0-0 R(3) to R(9) (with wavelengths 28.502, 23.034, 19.431, 16.894, 15.251, 13.593 and 12.472 µm, respectively). From their Einstein coefficients, excitation energies and the general low abundance of deuterium in the ISM all these lines were expected to be weak and unfortunately, they seem to be severely blended with other stronger emission features making impossible to obtain any reliable determination of fluxes.

4.2 Atomic species

Hydrogen.
The search for atomic hydrogen bands in the IC 348 gas results on the possible presence of the hydrogen band at 32.378 µm (see the HITRAN synthe as part of a blended emission feature at 32.40 µm. This emission feature has peak strength of 0.04 Jy relative to the continuum and integrated flux of $6.6 \times 10^{-18}$ W m$^{-2}$. Other hydrogen bands at 28.34 and 30.21 µm are either not detected or blended with stronger bands that make impossible to assess their presence in the observed spectrum. The hydrogen recombination lines H7α (HI 8-7) 19.062 µm, H6α 12.372 µm, H7β 11.309 µm and H8γ 12.385 µm are not detected. It is noted a strong emission line at 12.36 µm but the wavelength difference with H6α appears too large to support an association. The recombination line HI 13-9 at 14.183 µm could be contributing to a strong line (peak flux 0.035 Jy) seen at 14.175 µm. No evidence is found for any other bands of atomic hydrogen in the 10-30 µm spectral range.

Sulphur.
A search for the atomic sulphur [SI] $^3P_1 \rightarrow ^3P_2$ 25.249 µm line resulted in no detectable feature with strength above 0.01 Jy. The line at [S III] 18.71 µm is possibly detected with peak flux of 0.007 Jy, however the line at 33.48 µm is not detected (peak flux below 0.006 Jy). The ratio [SII]18.71/ [S III] 33.48 is a density diagnostic. CLOUDY models (Ferland et al. 1998) show that a ratio $>0.6$ corresponds to a density of $>100$ cm$^{-3}$.

Iron.
The well known [Fe II] 25.988 µm band could be responsible of an emission feature at 25.99 µm (peak flux of 0.03 Jy). However, the 24.519 µm and the 17.94 µm [Fe II] bands are not detected in the spectrum.

Neon.
The [Ne II] 12.81 µm bands is possibly present in the spectrum but it coincides with an OH and C$_2$H$_2$ 12.806 µm band. Given the spectral resolution of our spectrum it is not possible to separate the relative contributions of these bands. The ionization potential of this Ne emission line is 21 eV
and thus it would indicate that the slits may have captured emission from an HII region. The [Ne III] line at 15.56 \( \mu m \) is also seen in emission with a flux peak of order 0.03 Jy (excitation energy of the upper level is approx. 940 K). The [Ne V] line at 14.32 \( \mu m \) is however not present.

**Oxygen.**
The [O IV] line at 25.89 \( \mu m \) could be present in the IC 348 spectrum albeit with a peak flux below 0.007 Jy, only slightly above the detection limit.

In summary, the presence of weak bands of ionized atomic species of sulphur, iron and neon suggests that the observed interstellar gas includes the dominant component of warm gas and a region with relatively high excitation temperature.

### 4.3 Diatomic molecules

Absorption spectra of diatomic molecules commonly reported in the literature were computed using the python package HAPI and HITRAN. Fig. 5a and 5b show the synthetic spectra for a temperature of 250 K, adopted as representative of the gas in IC 348. A search for the strongest bands of each molecule was performed in the observed IC 348 spectrum. If the result was positive, then a more detailed analysis of weaker bands of that molecular species was carried out.

#### 4.3.1 OH Hydroxile

The relative strength of the OH bands in the 10-30 \( \mu m \) spectral region is quite sensitive to temperature (see Figure 6). Below 250 K the strongest band appears at 28.94 \( \mu m \), and also there are relatively strong doublets at 30.28, 30.35 and 30.66, 30.70 \( \mu m \). None of these bands are clearly detected in the spectrum of IC 348, neither the weaker bands predicted at shorter wavelengths.

We detect however a sequence of highly excited OH \((v = 0, J \rightarrow J' = 1)\) pure rotational transitions arising in the \(^2\Pi_{3/2} \rightarrow ^2\Pi_{1/2}\) rotational ladders (see wavelengths in Table 3 of Najita et al. 2010) which originate from high energy levels and have large transition probabilities (A-values of 20-70 s\(^{-1}\)). The bands more clearly seen in our spectra, depicted in Figures 7 and 8, correspond to high \(J\) values in the range \(J=29.5\) to \(J=20.5\) (with upper energy levels up to 4,000 K). Lines with lower \(J\) values appear weak or blended with other emission features in the spectrum.

It is unlikely that the chemical formation route via \(H_2 + O \rightarrow OH + H\) (e.g. Hollenbach & McKee 1979; Neufeld & Dalgarno 1989) is the dominant source of OH. This reaction has Arrhenius activation energies \(E_A/ k_B \) about 3150 K and only proceed efficiently in dense, hot gas. UV induced photodesorption of water ice from grain mantles and photodissociation of \(H_2O\) either in the gas phase or directly in the grain ice mantles (see Andersson et al. 2006, Tappe et al. 2008) could be the primary sources of OH in the IC 348 gas.

#### 4.3.2 CO
Carbon monoxide (CO) is the second most abundant molecule in the gas phase of the interstellar medium. In dense molecular clouds, it is also present in the solid phase as a constituent of the mixed water-dominated ices covering dust grains. Some rotational lines appear in our HITRAN based computed absorption spectrum, the most prominent are R (75), R (76) and R (77) at 35.51, 35.09 and 34.66 µm, respectively. These bands could be contributing to emission features in the IC 348 gas spectrum, but they are severely blended and it is not possible to ascertain their relative contribution against other contaminating bands. No other CO bands seem to be present in the IC 348 spectrum.

4.3.3 N$_2$

Isolated N$_2$ molecules are IR-inactive, and solid N$_2$ has only weak IR activity, hindering its astronomical detection by IR methods. The discovery of a new N$_2$ IR feature would be of great interest, the HITRAN synthetic spectrum shows that the strongest features appear at 11-11.5, 13.11, 15.46, 17.23, 21.84 and 22.83 µm. None of these bands could be unambiguously detected in our spectrum. The series of transitions between 11 and 11.5 µm are masked by the strong PAH 11.3 µm emission. The 13.11 µm band is barely resolved from the OH line at 13.08 µm, the 15.46 µm is blended and very weak with peak flux below 0.006 Jy, thus unreliable, the 17.23 µm line, if present, would be a small contribution blended with a relatively strong H$_2$O 17.225 µm water line (expected from the computations for water, see below). The lack of any emission at 21.84 µm (the strongest N$_2$ expected line) makes very unlikely that N$_2$ is causing any relevant contamination in the spectrum.

4.4 Tri-atomic molecules

Figure 9 shows HITRAN based computations in the range 10-19.5 µm for some of the most relevant triatomic molecules detected in diverse astrophysical environments: H$_2$O, CO$_2$, HCN, N$_2$O. In the spectral range 20-32 µm only water presents significant bands.

4.4.1 H$_2$O

Water has dipole-allowed pure rotational transitions that occur at mid/far-infrared and submillimeter wavelengths which have been extensively reported (Liseau et al. 1996, Harwit et al. 1998, Wright et al. 2000). In star-forming regions water is ubiquitous (e.g. van Dishoeck et al. 2021) with variations in its abundance relative to H$_2$ ranging from $< 10^{-8}$ in the coldest clouds to $> 10^{-4}$ in warm gas and shocks. The HITRAN computation for a temperature of 250 K shows a large number of water transitions in our spectral range. In Fig. 10a and 10b, the dependence with temperature is displayed. Many of these transitions have been previously observed in astrophysical environments (e.g. Pontoppidan et al. 2010, Blevins et al. 2016) and are also detected in our IC 348 gas spectrum with fluxes listed in Table 5. Fig. 11 shows water synthetic bands as compared with the observed spectrum in the range 12-13 µm. No evidence for significant water contamination is found in this spectral range. Water bands at 28.591, 22.639, 22.538, 17.358, 17.225 and 17.103 µm are detected (see Fig. 12). The bands at 30.525 and 30.529 µm are however not detected. At the moderate resolution of our spectrum, some of the water line complexes are severely blended rendering impossible to measure fluxes for individual transitions. When it was possible, band fluxes were calculated by defining and subtracting a linear fit to the local continuum
and fitting Gaussians to the lines. Lines with peak fluxes of less than 0.007 Jy are considered below the detection limit.

The water rotational diagram (Fig. 13) was built with the observed fluxes and the molecular parameters listed in Table 5. It is derived an excitation temperature of T=380 K and a column density $N_{\text{tot}}(\text{H}_2\text{O}) = 2.7 \times 10^{15}$ cm$^{-2}$. The $[\text{N(H}_2\text{O)}]/[\text{N(H}_2)] \sim 10^{-5}$ is not unusual in regions with warm gas and shocks (van Dishoeck et al. 2021).

While there is a significant presence of water bands in our spectrum, the many high excitation temperature transitions $> 4000$ K of water expected in our spectral range appear too weak for being detected at the resolution and sensitivity of the present spectrum, and therefore do not cause a significant problem of contamination for the identification of amino acid bands.

4.4.2 CO$_2$

The strongest CO$_2$ band in our spectral range as it can be seen in Fig. 14 is the well known $\nu_2$ 1–0 14.98 µm band. Detections of this band are extensively reported in the literature (van Dishoeck et al. 1996, van Dishoeck 1998, Dartois et al. 1998, Boonman et al. 2003) in a variety of star-forming regions. The CO$_2$ abundances with respect to H$_2$ obtained are typically $2 \times 10^{-7}$, up to two orders of magnitude lower than those of solid CO$_2$ in the same regions.

The 14.98 µm band is clearly seen in the IC 348 spectrum with a peak flux of 0.017 Jy with respect to the local continuum. The other CO$_2$ bands in the computed spectrum are significantly weaker and thus expected with peak fluxes below 0.01 Jy. Some of these bands could indeed be present, i.e. the doublet at 13.876, 13.898 µm at peak flux about 0.007 Jy, close to the detection limit. Many weak CO$_2$ lines predicted by the HITRAN computation, are likely contributing to the confusion level at fluxes of order 0.005 Jy. These individual lines cannot be resolved or identified at the resolution and sensitivity of our spectrum.

4.4.3 HCN Hydrogen cyanide

The HITRAN computation displays a rich HCN spectrum in the wavelength range of our interest (see Fig. 15) with the strongest band being the $\nu_2$ 1-0 fundamental ro-vibrational band at 14.05 µm (see e.g. Lahuis and van Dishoeck 2000). We find in the IC 348 spectrum a relatively strong feature (0.01 Jy peak flux) at 14.04 µm which could be associated to this HCN band (blend of transitions $12_{76} \rightarrow 11_{47}$ and $10_{73} \rightarrow 9_{46}$). In Fig. 15 we can see the comparison between the synthetic and the observed spectrum. The next most intense HCN band according to the computation may contribute to the emission feature seen at 13.977 µm. However, given the relative strength with respect to the 14.05 µm band, its contribution to this feature should be minor. The weaker HCN feature at 13.76 µm seems to be present and well resolved in the IC 348 spectrum with a rather low peak flux of 0.007 Jy. Other HCN bands may also be present at similar strength, i.e close to the detection limit. At such small flux level all of them appear blended with other emission features and do not represent a major problem for the identification of amino acids.
Hydrogen isocyanide HNC is expected to be less abundant than HCN, the strongest HNC ν2 band at 21.54 µm is not detected in the IC 348 spectrum.

4.4.4 NO2

The HITRAN computation shows the strongest band of this molecule is located at 12.65 µm. At this wavelength, there is a very weak feature (noise level in the IC 348 spectrum. Since the other bands in the synthesis are considerably weaker, no contribution is expected from this molecule to any emission lines above the detection limit.

4.4.5 N2O

The HITRAN computation shows the strongest band of this molecule is at 16.97 µm. This band is below the detection level in the IC 348 spectrum. The remaining synthetic bands are considerably weaker, thus no significant contribution to any emission feature is expected from this molecule.

4.5 Tetra-atomic molecules

Fig. 16 shows the synthetic spectra obtained with HITRAN for C2H2, NH3 and SO3.

4.5.1 C2H2  Acetylene

C2H2, a well known precursor of PAHs, and other symmetric hydrocarbons is predicted to be quite abundant in the interstellar medium. These symmetric hydrocarbons do have permitted infrared vibration-rotation transitions. The ν5 band has three branches, P(J-1), Q(J), R(J+1) where J is the rotational quantum number of the upper vibrational state. The ν5 band 9 5 5 → 8 0 8 + 14 7 8 → 13 49 at 13.71 µm is the strongest ro-vibrational band of C2H2 (symmetric bending mode in which the two H atoms vibrate together) detected in diverse astrophysical environments (Lacy et al. 1998, Lahuis and van Dishoeck 2000, Boonman et al. 2003, Carr and Najita 2008, Najita et al. 2021). A well resolved emission feature in the IC 348 spectrum with a peak flux of 0.008 Jy with respect the local continuum is found at 13.71 µm (see Fig. 17) and could be caused by this C2H2 band. The measured flux of this band is 2 \times 10^{-18} W m^{-2}. If we assume this molecule is at the same temperature than H2 it would lead to a column density of N(C2H2) of order 1 \times 10^{12} cm^{-2}. The R(23) C2H2 at 12.73 micron could also be present, blended with the R(24) HCN line at the same wavelength. Other C2H2 bands in the HITRAN-based computation are predicted much less intense than these two bands, thus below the detection limit of our spectrum.

4.5.2 NH3

NH3 bands reported in the mid-IR (see e.g Najita et al. 2021) were searched in the IC 348 spectrum. The 10.74-10.76 µm emission feature (peak flux 0.01 Jy, see Fig. 18) can be associated to NH3 antisymmetric transitions aQ(3,3), aQ(4,4), aQ(7,6). In addition, evidence for NH3 bands
at 10.50 and 10.35 µm is also found as weak emission features (with peak flux 0.007 Jy and 0.005 Jy, respectively).

4.5.3 CH₃

The methyl radical CH₃ is an important intermediate product in the basic ion-molecule gas-phase chemistry networks in the interstellar medium driven by cosmic-ray ionization (Herbst & Klemperer 1973) and it is an important building block in the formation of small hydrocarbon molecules. Together with CH and CH₂, it is produced by a series of reactions starting with \( \text{C} + \text{H}_3^+ \rightarrow \text{CH}^+ + \text{H} \) or via the radiative association of \( \text{C}^+ + \text{H}_2 \rightarrow \text{CH}_2 + \text{hv} \), followed by hydrogen abstraction reactions and dissociative recombination. Alternatively, CH₃ can be produced by the photodissociation of methane (CH₄). Subsequent reactions of \( \text{C}^+ \) with CH₃ form one of the most important steps in the formation of small hydrocarbons. It is interesting to search CH, CH₄ and C₂H₂ along the same line of sight as it traces basic gas-phase astrochemistry networks. CH was reported by Snow et al. (1994) in the line of sight towards the center of IC 348. N(CH)=5.3 x 10^{13} \text{ cm}^{-2} \) and C₂H₂ was discussed above. Here we present evidence for CH₃ in the gas of IC 348.

The strongest CH₃ Q–branch transition is located at 16.5 µm; the other strong feature is the R (0) line at 16.0 µm thanks to its favorable Hönl-London factor. The 16.0 and 16.5 µm (ν₂ bending mode bands) were detected by Feuchtgruber et al. (2000), toward the Galactic Center Saggitarius A with column density of 8.05 (+/-2.4) x 10^{14} \text{ cm}^{-2}.

At 16.00 µm there is an emission feature in the IC 348 spectrum which also may coincide with a band of Tryptophan. At 16.52 µm there is another weak feature which could be due to CH₃, if so, it implies also a minor contribution of this molecule to the 16 µm band. R(1) is at 15.54 µm could be present as a weak blue shoulder in the strong emission feature at 15.56 µm previously discussed as being caused by a combination of [Ne III] and H₂O and the P(2) at 17.60 µ is below the detection limit.

4.6 Polyatomic Molecules

4.6.1 Five atoms

CH₄ Methane

Lacy et al. (1991) detected the CH₄ R(0) and R(2) lines with ground based observations toward NGC 7538 and Dartois et al. (1998) and (Boogert et al. 1998) report observations of gaseous and solid phase CH₄ toward the embedded protostar GL 7009S. These are bands at wavelengths shorter than covered by our spectrum. There are no relevant bands of CH₄ in the IC 348 spectrum.

HC₃N Cyanoacetylene

This molecule is one of the building blocks from which more complex hydrocarbons are produced.
(Cernicharo et al. 2001). The strongest band of HC₅N is ν₅ located at 15.10 µm (see the HITRAN based synthetic spectrum in Fig. 19 (Yamada & Bürger 1986) could be responsible of a weak emission feature found at 15.11 µm in the IC 348 spectrum with peak flux of 0.007 Jy.

4.6.2 Six atoms

**C₂H₄ Ethylene**

There is a clear emission feature at 10.53 µm (peak flux 0.011 Jy) which may be caused by the 10.53 µm Q branch of the ν₇ CH₂ wagging-mode of the C₂H₄ (Cernicharo et al. 2001, Bast et al. 2013). The strongest line in the HITRAN computation is located at 15.9 µm, as we will see below this is also closely located to the strongest line of C₄H₂, and both lines could be contributing to the broad but weak emission feature at 15.84-15.90 µm seen in the IC 348 spectrum with a peak flux of order 0.01 Jy.

**C₄H₂ Diacetylene**

Cernicharo et al. (2001) reported the detection of C₄H₂ in ISO observations of CRL 618. The absorption signal near 15.9 µm was identified and assigned to the ν₈ 15.87 µm fundamental bending mode of C₄H₂ based on the laboratory work of Arié & Johns (1992). In the IC 348 spectrum there is an emission feature at 15.87 µm with peak flux of 0.01 Jy, but it is severely blended with other emission bands of similar strength (see Fig. 20).

4.6.3 Seven atoms

**CH₃C₂H propyne or methylacetylene**

It has its strongest band at 15.79 µm. No emission feature could be found at this wavelength.

**HC₅N**

It is expected that HC₅N (like HC₃N) is formed by the reaction of C₄H₂ + CN. As we may have found evidence for C₄H₂ it is appropriate to search for HC₅N. According to Deguchi and Uyumura (1984), the strongest transition of this molecule in our spectral range (see Fig.20) is ν₇ at 14.60 µm (685 cm⁻¹), followed in strength by the ν₈ 17.67 µm (566 cm⁻¹). Both lines seem to be present in the spectrum of the interstellar gas of IC 348, albeit rather weak (peak flux of 0007 Jy) and possibly blended with other similarly weak lines.

4.6.4 Eight atoms

**C₂H₆ Ethane**

C₂H₆ is an important molecule in planetary and cometary atmospheres (see e.g. Burgdorf et al.

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**Note:** The text continues with more details on the detection and properties of various molecules and their interactions in astronomical contexts, focusing on specific wavelengths and line strengths observed in interstellar and cometary spectra.
2006, Coustenis et al. 2003). It has strong rotational-vibrational bands in the infrared, and the \( \nu_9 \) band at 12 \( \mu m \) has been widely studied (Daunt et al. 1981, Vander Awera et al. 2006).

The HITRAN computation shows a broad system of lines associated to the \( \nu_9 \) mode with the strongest transition at 12.16 \( \mu m \) (822.35 cm\(^{-1}\)). A relatively broad feature (0.03 \( \mu m \)) with peak intensity 0.009 Jy is detected at 12.15 \( \mu m \) in the IC 348 spectrum which could be due to this \( \nu_9 \) band of \( C_2H_6 \).

\( C_6H_2 \) triacetylene

According to laboratory work (e.g. Haas et al. 1994) the \( \nu_{11} \) fundamental bending mode of \( C_6H_2 \) is produced at 16.10 \( \mu m \). Cernicharo et al. (2001) reported the detection of this band in ISO observations of CRL 618. In the IC 348 gas spectrum there is a clear well resolved emission feature at 16.10 \( \mu m \) which we ascribe to \( C_6H_2 \) (see Fig. 20). The measured flux of the 14.84 \( \mu m \) emission feature mainly attributed to benzene is 4 (±0.5) \( \times 10^{-18} \) W m\(^{-2}\).

**4.6.5 More than 9 atoms.**

\( C_6H_6 \) Benzene

The bending mode \( \nu_4 \) (all hydrogens moving away in the same direction from the plane formed by the six carbons) is the strongest infrared band of \( C_6H_6 \). The lines in the \( \nu_4 \) Q-branch are overlapped, producing a relatively strong narrow feature at 14.837 \( \mu m \) (Lindemayer 1988, Cernicharo et al. 2001). In addition to the \( \nu_4 \) transition, there is a hot band at 14.862 \( \mu m \), \( \nu_4+\nu_{20} \rightarrow \nu_{20} \) where \( \nu_{20} \) is the lowest frequency vibrational mode of benzene at 398 cm\(^{-1}\). The vibrational band strength for the hot band could be similar to that of the fundamental \( \nu_4 \) (see Kauppinen, Jensen, & Brodersen 1980). In the IC 348 spectrum there is a clear emission feature at 14.84 \( \mu m \) (with peak flux of order 0.013 Jy) which may have significant contributions from the \( \nu_4 \) and hot \( C_6H_6 \) bands and also from the P(13) line of HCN at 14.847 \( \mu m \). The measured flux of the 14.84 \( \mu m \) emission feature (Fig. 21) mainly attributed to benzene is 4 (±0.5) \( \times 10^{-18} \) W m\(^{-2}\).

The detection of \( C_6H_6 \) and other small molecular species like \( C_4H_2 \), \( C_6H_2 \), HC5N suggest that the conditions in the gas of IC 348 are adequate to activate a rich and diverse chemistry which may have produced more complex hydrocarbons.

**4.7 PAHs**

The C-C and C-H stretching and bending modes of polycyclic aromatic hydrocarbons (PAHs) at 6.2, 7.7, 8.6, 11.3, 14.2, 16.2 \( \mu m \) dominate the mid-IR spectra of many objects and are indicators of the presence of complex carbonaceous material excited by UV radiation (Leger & Puget 1984, Allamandola et al. 1985).

The PAH transitions in our range of interest 11.3, 12.0, 12.7, 14.2, 16.2 microns correspond to C-C modes. Particularly strong in the IC 348 spectrum is the PAH transition at 11.3 \( \mu m \) micron with peak flux of 0.05 Jy. The other transitions are weaker but also present as relatively broad emission
features of 0.1-0.2 micron width to which other narrow emission bands are superposed. The carriers emitting at mid-IR are expected to be small molecules of order 10 Å which correspond to PAHs with 30 to a few hundred carbon atoms (e.g. Boulanger et al. 2000).

4.8 Fullerenes

The detection and determination of fullerene abundances in various interstellar regions of IC 348 was reported by Iglesias-Groth (2019) using Spitzer IRS data. Several bands of C\textsubscript{60}, C\textsubscript{70} and C\textsuperscript{+}\textsubscript{60} were identified. The spectrum under study here involves various other regions of the interstellar medium in IC 348 and several emission features are also found consistent with known fullerene bands (peak fluxes are given in parenthesis), most remarkably: for C\textsubscript{60} 17.33 \(\mu\)m (0.02 Jy) and 18.88 \(\mu\)m (0.06 Jy); for C\textsubscript{70} 12.63 \(\mu\)m (0.01 Jy), 13.83 \(\mu\)m (blended), 14.90 \(\mu\)m (0.012 Jy), 15.63 \(\mu\)m (0.03 Jy), 17.78 \(\mu\)m (0.035 Jy) and, for C\textsuperscript{+}\textsubscript{60} 10.46 (0.015 Jy), 13.22 (0.012 Jy) and 18.58 \(\mu\)m (0.03 Jy).

4.9 Ices

Water was the first molecule detected in the solid state in the interstellar medium (Gillet and Forrest 1973). Since then many other molecules have been identified in icy form (i.e. CO, CO\textsubscript{2}, CH\textsubscript{4}, NH\textsubscript{3} and CH\textsubscript{3}OH). These ices are known to be common during the cold and dense stages of star formation, with abundances reaching 10\textsuperscript{-4} n(H\textsubscript{2}). Icy dust grains play a key role in the formation of more complex organic molecules (COMs), such as glycolaldehyde (HOCH\textsubscript{2}CHO) and ethylene glycol (HOCH\textsubscript{2}CH\textsubscript{2}OH).

H\textsubscript{2}O.
The most direct evidence for solid-state formation of interstellar water comes from the detection of the 3 \(\mu\)m O-H stretching vibration band of water ice toward numerous infrared sources. In many cases, the 6 \(\mu\)m and 11 \(\mu\)m librational modes have also been observed. Giuliano et al. (2014) presents laboratory spectra of amorphous and crystalline water in the spectral range of our interest, both spectra display 10-20 \(\mu\)m wide bands which are not seen in our spectrum of the gas in IC 348.

CO\textsubscript{2}

Based on laboratory spectroscopy of mixed CO\textsubscript{2} ices (D’Hendecourt & Allamandola 1986) the \(\nu_2\) bending mode of CO\textsubscript{2} ice in absorption at 15.20 \(\mu\)m was identified by D’Hendecourt and Jourdain de Muizon (1989) toward several sources in the IRAS database. Ehrenfreund et al. 1997 showed the existence of new bands associated to this bending mode when CO\textsubscript{2} is present in a mixture of other ices. Multiple laboratory experiments measured the shifts and the presence of various bands as a function of the mixture. In several cases three bands of similar strength were found around 15.2 \(\mu\)m. In the IC 348 gas spectrum we find three strong well resolved emission features in this region at 662.7 cm\textsuperscript{-1}, 660.5 cm\textsuperscript{-1}, y 657.5 cm\textsuperscript{-1} which fit well CO\textsubscript{2} ice bands in the case of a mixture of CO\textsubscript{2}:CO\textsubscript{2} (see table 3 in Ehrenfreund et al 1997). There may be other mixtures that could also reproduce these features, so this is only a possible explanation for the
bands detected between 15.0 and 15.2 µm.

**NH₃**

Giuliano et al. (2014) give measurements of NH₃ ices and other ice transitions in our spectral region. For NH₃ the reported bands are at 439 cm⁻¹ (23.87 µm) for amorphous ice, and 535 and 421 cm⁻¹ for crystalline ice (at 18.69 µm and 23.75 µm, respectively). A weak band in the IC 348 spectrum at 23.87 µm could be due to amorphous NH₃ ice. The crystalline bands are undetected or severely blended with bands of other species.

**CH₃OH**

CH₃OH and other ices are proposed sources of complex organic molecules (Charnley et al. 1992; Garrod et al. 2008). The determination of ice abundances and production channels in star-forming regions is very relevant for studies of prebiotic chemistry. Hudgins et al. (1993) measured the ν₁₂ mode of CH₃OH at 705 cm⁻¹ (14.184 µm). An emission feature at 14.175 µm in the spectrum of IC 348 could be associated to this band. The amorphous CH₃OH 330 cm⁻¹ (30.30 µm) band could be associated to a rather strong feature in the IC 348 spectrum with peak flux 0.04 Jy. The crystalline 347 cm⁻¹ band at 28.82 µm is possibly detected, although with a modest peak flux of 0.008 Jy.

**5. AMINO ACIDS RESULTS**

Laboratory wavenumbers, wavelengths (laboratory and observations) and integrated molar absorptivities (ψ) for the strongest mid-IR transitions of the amino acids under study are listed in Table 6. Laboratory wavenumbers are precise at the level of 1 cm⁻¹ and integrated molar absorptivities (adopted from Iglesias-Groth and Cataldo 2021) have uncertainties of order 20%. In the Table it is noted whenever a proposed amino acid band may be blended with any of the bands of the molecules discussed in the previous section. In Figures 3-8 the positions of the strongest laboratory bands of each of the five amino acids under study and the emission features we tentatively assigne to them are marked on the combined IC 348 ISM spectrum (red-line). For comparison, in each figure it is also plotted the star-forming region ISM “reference” spectrum (black line). For all the strongest mid-IR bands of amino acids we have identified potential emission features in both spectra with wavelengths fully consistent within errors with those measured at laboratory.

Both, the IC 348 and the reference ISM spectra show a very large number of emission lines in common. The most intense lines are due to well identified molecular and atomic transitions. The strongest and broader feature in the Perseus spectrum is the 11.2 µm PAH band. Other strong lines which we can see in the panels of Figures 22, 23 and 24 are associated to transitions of molecular hydrogen at 12.28 and 17.04 µm, and atomic hydrogen at 12.37 and 19.06 µm. Weaker lines of water (marked with blue asterisks), fullerene species and other organic molecules are also seen in the spectra as discussed above but only cause contamination to some of the proposed amino acid bands.

Fluxes for the assigned amino acid bands were measured only in the IC 348 ISM spectrum by integrating the continuum subtracted spectrum. In Table 6 we list, for each of the five amino acids,
measured fluxes and wavelengths for all the laboratory bands with integrated molar absorptivities \( \psi > 2 \text{ km mol}^{-1} \). Errors in the band fluxes were estimated by propagating pixel error through the band integrals and are typically of order 20%. We tentatively identify 14 lines for tryptophan, 12 lines for tyrosine, 7 lines for phenylalanine and 5 lines for isoleucine and glycine, respectively. For all the strongest laboratory bands we find a counterpart emission line in the observed spectrum at relative strengths consistent with those measured in the lab.

Tryptophan. In Table 6 there are 12 bands with reported integrated molar absorptivities \( \psi > 2 \text{ km mol}^{-1} \), we identify in Fig. 22 emission features which could be associated to each of these laboratory bands and provide fluxes for each of them. We also list fluxes for emission lines that coincide with other reported weak bands of tryptophan for which there are no available molar absorptivities. The two bands with larger integrated molar absorptivity \( (\psi >30 \text{ km mol}^{-1}) \) have clear counterparts at 13.45 and 19.64 \( \mu \text{m} \). There are also four laboratory bands with \( \psi >10 \text{ km mol}^{-1} \) which coincide with emission features in the spectra at 11.58, 17.88, 23.47 and 28.70 \( \mu \text{m} \). Other bands with low values of \( \psi = 2-5 \text{ km mol}^{-1} \) seem to be present in the spectrum albeit at more modest fluxes. For any reported laboratory band with a measured \( \psi \) we find a counterpart emission feature in the observed spectrum as marked in the figure.

Tyrosine. All the 12 bands listed in Table 6 have a counterpart in the observed spectrum (see Fig. 23). There are six bands with \( \psi > 10 \text{ km mol}^{-1} \) but two of them at 17.36 and 18.86 \( \mu \text{m} \) are not plotted because they are severely blended with well known water and fullerene bands. The band at 26.32 \( \mu \text{m} \) is marginally resolved (the observed feature is too broad) and very likely contaminated by an unknown specie at a slightly longer wavelength. The other six remaining weak bands of tyrosine have their proposed counterparts marked in Fig. 23 and fluxes are reported in the table.

Phenylalanine. All eight lines listed in Table 6 have tentative counterparts marked in Fig. 24. The bands with the two higher \( \psi \) values \( (\psi >20 \text{ km mol}^{-1}) \) at 14.29 and 27.31 \( \mu \text{m} \) also have the higher measured fluxes. The line at 27.31 \( (\psi >50 \text{ km mol}^{-1}) \) appears however with a red tail which indicates some contamination. The line at 14.29 \( \mu \text{m} \) is weaker but with a well defined profile. The other bands have lower \( \psi \) values and also the assigned lines in the spectrum present lower fluxes.

Isoleucine. The eight bands listed in Table 6 have low \( \psi \) values with the highest, \( \psi = 13 \text{ km mol}^{-1} \), corresponding to the 18.58 \( \mu \text{m} \) band which also shows the highest observed flux in the proposed counterpart emission features. Three of the other seven bands are clearly seen in Fig. 25 but are unfortunately blended with other molecules and two of the remaining bands have low fluxes at only 2 sigma from the detectability limit making the evidence for this amino acid is less solid than for the previous ones.

Glycine. There are only six known bands in the spectral range under consideration, out of which five have integrated molar absorptivities reported (see Fig. 26). The two bands with higher \( \psi \) values at 19.85 and 27.75 \( \mu \text{m} \) and the 14.32 \( \mu \text{m} \) band can be assigned to rather isolated emission lines in the IC 348 spectrum. The band at 10.2 \( \mu \text{m} \) is blended with the strong PAH band and its presence (and flux) is difficult to ascertain. The remaining band at 16.45 \( \mu \text{m} \) could be associated with an emission line which is superimposed on a weak broader spectral feature and the measured flux is rather uncertain. The evidence for the presence of glycine in the ISM of IC 348 relies on the proper
identification of only a few available bands and shall be taken with caution.

The bands with the largest laboratory integrated absorptivities generally show stronger emisión lines counterpart in the observed spectra, both in Perseus and in the reference spectrum. Emission features likely contaminated by water lines are indicated in Table 6, but we caution that emission features assigned to amino acids may also be contaminated by other unknown species. Future high resolution mid-IR observations should clarify the extent of this contamination. The vibrational excitation diagrams (see below) suggest that if contamination exists in some bands, it is not a dominant contribution to the flux.

5. DISCUSSION

5.1 Exploring excitation diagrams and equilibrium temperatures

In order to produce vibrational excitation diagrams it is necessary to estimate for each transition the number of molecules in the upper vibrational state \( N_u \). This can be obtained from the flux of each transition assuming optically thin emission. The total power emitted in a band (see e.g. Camí et al. 2010) is

\[
P = N_u A_{ul} h \nu_{ul} / 4\pi D^2,
\]

where, \( D \) is the distance to IC 348 (321 pc), \( \nu_{ul} \) the frequency of the amino acid transition, and \( h \) the Planck constant. The Einstein \( A_{ul} \) coefficients can be obtained from the laboratory integrated molar absorptivities, which ideally should be measured in conditions resembling those of the astrophysical environment under study. As molar absorptivity coefficients are sensitive to temperature and to vacuum conditions, we only expect from the available absorptivity data a very preliminary indication of the existence of thermal equilibrium.

Vibration excitation diagrams for each amino acid were obtained using the strongest transitions for which molar absorptivities were available and fluxes measured in the “combined IC 348 ISM” spectrum (listed in Table 6). The results for each individual amino acid, plotted in Figure 27, show good correlation coefficients in the range \( r=0.9-0.95 \), even if a vibrational degeneracy \( g_u=1 \) was adopted for all the energy levels given the lack of information in the literature. The inverse of the slopes of the fits in the figure indicates equilibrium temperatures in the range 270-290 K for each amino acid. This is remarkably similar to the excitation temperature we found for H\(_2\). Fortunately, this is also very close to the temperature of the laboratory setting used for the measurement of the integrated molar absorptivities. The diagram suggests that amino acids exist in rather warm gas of the stellar cluster. Values for total number of emitting molecules of amino acids are found in the range \( N_{tot}=10^{42} \) from the ordinate at origin, this shall be taken only as order of magnitude indicative given the various approximations and limitations involved. For comparison, fullerene \( C_{60} \) and \( C_{70} \) estimates in IC 348 based on Spitzer spectra obtained with the same instrument at similar locations in the core of the IC 348 cluster led to equilibrium temperature estimates of order 200 K and \( N_{tot} \) values about one hundred times higher (Iglesias-Groth 2019).

5.2 Column densities

Assuming that amino acids are present in the ISM of IC 348 we may attempt to estimate their column densities. Considering that the absorbed UV energy is re-emitted mostly via the IR
vibrational bands, column densities, n(AA), can be derived from the measured band fluxes in the IC 348 spectrum. Reliable absolute abundances would require molar absorptivities of the mid-IR bands measured in conditions resembling the ISM in Perseus. The available laboratory data may not be ideal, but can suffice to provide useful insight on the relative abundances of the amino acids under consideration. We will follow procedures similar to those used to estimate abundances for other molecular species in the ISM of star-forming regions (e.g. Berné et al. 2017, Iglesias-Groth 2019). The total IR intensity (W m\(^{-2}\) sr\(^{-1}\)) emitted by the amino acid AA can be estimated as \(I_{\text{tot}} = n(AA) \times \sigma_{\text{UV}} \times G_0 \times 1.2 \times 10^{-7}\) where \(G_0\) is the radiation field in the locations of the observations and \(\sigma_{\text{UV}}\) is the cross section for absorption in the UV of the relevant molecular specie.

The standard value of the radiation field in the general ISM (Habing 1968), \(G_0=1\), corresponds to \(1.2 \times 10^{-7}\) W m\(^{-2}\) sr\(^{-1}\). Large variations exist in the far UV radiation field within the IC 348 star-forming region (Habing 1968). As the location of the observations under consideration are relatively close to the positions of the most luminous stars of the cluster, the appropriate value of \(G_0\) will be significantly higher. Previous work on fullerene abundances using spectra obtained in very similar locations of the ISM in IC 348, adopted \(G_0 = 45\) for the average interstellar radiation field (Iglesias-Groth 2019). The same value will be adopted here.

The total intensity, \(I_{\text{tot}}\), can be determined from the measured total fluxes emitted in the IR. However, we only measured fluxes for the subset of stronger lines known in the 10-30 \(\mu\)m region. The true total IR intensity will be higher than the value inferred from our band measurements, and subsequently, the column densities too. The excitation diagram suggests that amino acids in the ISM of IC 348 are in thermal equilibrium with excitation temperatures in the range 270-290 K (Fig. 27). We will assume such thermal equilibrium conditions to model the contribution to the total IR flux resulting from the IR transitions out of the range 10-30 \(\mu\)m for which molar absorptivities are also available (Iglesias-Groth and Cataldo 2021) and then, apply an upward correction to the total flux for each molecular specie. The resulting correction factors are 2 for tryptophan, 3 for tyrosine and isoleucine and 4 for glycine and phenylalanine. The estimated total emitted IR fluxes after applying these corrections are 10.6, 8.6 and 0.8 \(\times 10^{-17}\) W m\(^{-2}\) for the aromatic tyrosine, phenylalanine and tryptophan respectively, and 5.2 and 12.9 \(\times 10^{-17}\) W m\(^{-2}\) for the aliphatic isoleucine and glycine, respectively. As we have used full aperture extraction spectra, band fluxes are converted into \(I_{\text{tot}}\) dividing by the subtended area in the sky by the corresponding slit of each module of the IRS spectrograph.

The third parameter needed to infer column densities is the amino acid UV absorption cross section, \(\sigma_{\text{UV}}\). This can be derived from molar absorptivities. The aromatic amino acids contain conjugated aromatic rings and therefore are very efficient absorbing light in the UV range. UV absorption cross sections \(\sigma(\text{cm}^2)\) can be computed from laboratory measurements of molar absorption coefficients \(\varepsilon(\lambda)\) using the relation \(\sigma(\lambda) = 1000 \varepsilon(\lambda)/(N_A \log(e))\) where \(N_A\) is the Avogadro number, and \(\log(e)\) is the decimal logarithm of the Euler number. In Fig. 28 we display molar absorption coefficients \(\varepsilon(\text{mol}^{-1} \text{cm}^{-1})\) for the five amino acids under consideration (Wetlaufer 1963, Fasman 1976). The plot shows molar absorption coefficient values as a function of wavelength for the range of interest 200-300 nm.

In the IC 348 star-forming region the most luminous stars dominating the UV spectrum are of type B5 and A2 with effective temperatures of 15000 and 10000 K, respectively, and the peak of their...
combined emitted radiation is located around 200 nm. At shorter wavelengths the flux emitted by these stars sharply decreases. More than 400 cooler low-mass stars have been identified (Luhman et al. 2016) in IC 348, but their luminosities are much lower and the peak of their emitted radiation occurs at wavelengths longer than 300 nm where the amino acids under study have negligible absorbances (see Figure 28). Thus, given the expected stellar radiation field the most relevant wavelength range for determining effective UV absorption cross sections is from 190 to 230 nm and the resulting effective molar extinction coefficients, \( \varepsilon_{\text{eff}} \) are 10850, 5640, 3530, 90 and 50 \( \text{mol}^{-1} \text{cm}^{-1} \), for tryptophan, tyrosine, phenylalanine, isoleucine and glycine, respectively. Note the very low molar extinction coefficients of glycine and isoleucine as compared with those of the aromatic amino acids. Adopting these values, we obtain for the corresponding mean UV absorption cross sections 4, 2, 1, 0.03 and 0.02 \( \times 10^{-17} \text{ cm}^2 \), for tryptophan, tyrosine, phenylalanine, isoleucine and glycine, respectively.

The resulting column densities for the aromatic amino acids are then: phenylalanine \( n(\text{Phe}) = 1 \times 10^{11} \text{ cm}^{-2} \), tyrosine \( n(\text{Tyr}) = 0.8 \times 10^{11} \text{ cm}^{-2} \), and tryptophan \( n(\text{Trp}) = 0.6 \times 10^{11} \text{ cm}^{-2} \). For isoleucine we find a much higher column density \( n(\text{Ile}) = 2 \times 10^{12} \text{ cm}^{-2} \). For glycine, the corresponding column density based on the few transitions available in our spectral range appears to be the highest \( n(\text{Gly}) = 9 \times 10^{12} \text{ cm}^{-2} \). We recall that estimates of the latter two amino acids relay on the identification of only a few bands for each of them and that the total IR fluxes are more uncertain than for the aromatic amino acids. Additional observations of glycine and isoleucine in Perseus using transitions in other spectral ranges would be highly valuable. An upper limit to the glycine column density of \( 5 \times 10^{12} \text{ cm}^{-2} \) was established for the low-mass protostar IRAS 16293-2422 (Ceccarelli et al. 2000) in a different star-forming region, it would be important to carry out a similar sensitive search for millimetric transitions of this amino acid across the Perseus Molecular Complex.

5.3 Relative abundances

Glycine is the most abundant amino acid in any type of carbonaceous chondrites with abundances of order 5-6 ppm in CM2 types and 200 ppm in CR2 types, the meteorites with a higher abundance of amino acids. In both types, isoleucine, phenylalanine and tyrosine are also systematically detected although at lower concentrations (Coob & Pudritz 2014), with abundance levels of order 1, 0.8 and 0.9 ppm, respectively, for CM2 types. In CR2 types, isoleucine is more abundant with values of order 30 ppm, followed by phenylalanine 20 ppm, while tyrosine is at the level 0.7 ppm. Among the amino acids studied in this work, glycine and isoleucine are the most abundant in CR2 meteorites. They appear to be also the most abundant in the ISM of the IC 348 star-forming region. On the other hand tryptophan, which we find to be the less abundant of the five studied amino acids in Perseus, interestingly, it is undetected in meteorites.

Another relevant comparison with meteorites is the fraction of gas phase locked carbon in amino acids. This can be estimated from the amino acid column densities via \( f_C(\text{AA}) = \frac{n(\text{AA}) \times N_{\text{aa}}}{N(\text{H}) \times [C]} \), where \([C]=1.6 \times 10^{-4}\) is the carbon to hydrogen ratio in the region (Sofia et al. 2004), \(N_{\text{aa}}\) is the number of carbon atoms in the amino acid and \(N(\text{H})\) is the hydrogen column density, \( 4.8 \times 10^{22} \text{ cm}^{-2} \) for the central part (Sun et al. 2008) of IC 348. The results are \( f_C = 0.08, 0.09, 0.1, 1.8 \)
and $2.4 \times 10^{-6}$ for tryptophan, tyrosine, phenylalanine, isoleucine and glycine, respectively. It is remarkable that these values for isoleucine and glycine are very close to the 3 parts per million versus carbon reported in several types of carbonaceous chondrites (Cronin & Chang 1993, Botha & Bada 2002).

The emission features assigned to amino acids in the ISM spectrum of Perseus are also present in the combined ISM spectrum from star-forming regions. Column densities cannot be inferred from this spectrum because the sources are distributed in a large range of distances and fluxes would not be meaningful, however the spectrum clearly displays the same bands we have assigned to amino acids with relative intensities similar to those found in IC 348. This may indicate that amino acids are widely distributed across the Galaxy.

6. CONCLUSIONS

We have conducted a search for mid-IR (10-30 µm) transitions of five amino acids (tryptophan, tyrosine, phenylalanine, glycine and isoleucine,) in the Spitzer spectrum obtained averaging observations from various ISM locations in the core of the IC 348 star forming cluster in the Perseus Molecular Cloud. Each of the strongest laboratory bands of these amino acids have a detected counterpart emission feature in this spectrum. We propose that amino acids are a likely explanation to the observed bands. A systematic search for possible contaminants to these bands was carried out and bands of several molecules were identified in the spectrum of IC 348 ($H_2$, $H_2O$, $CO_2$, $OH$, $C_2H_2$, $C_4H_2$, $C_6H_6$, PAHs, fullerenes). Only a small number of the proposed amino acid bands seem to have contamination from these molecular species. While we cannot discard that additional contamination from other less common molecular species it is possible, the proposed explanation in terms of amino acids remains valid.

Using the bands ascribed to amino acids we have built preliminary vibrational excitation diagrams from which equilibrium temperatures of amino acids are inferred in the range 270-290 K. This estimate should be taken with caution, but interestingly the derived temperatures are very close to those found for the much more abundant molecular hydrogen. From the available UV absorption cross sections and estimated total IR fluxes, we have estimated what would be the amino acid column densities and found that the relative abundance pattern of these molecules in the ISM of IC 348 is similar to that known in meteorites, with glycine and isoleucine being much more abundant than any of the other three aromatic amino acids. The fraction of gas phase locked carbon in amino acids in the ISM of IC 348 also appears to be similar to that reported for carbonaceous chondrites.

If amino acids are present in the ISM of star-forming regions, they could also be part of the inventory of organic molecules in protoplanetary discs. Searches in protostars and protoplanetary discs in Perseus and other molecular cloud complexes are worthwhile as they should provide valuable insight on the delivery of complex organics by meteoritic and cometary material to planets in early stages of formation and, ultimately, on the processes relevant to the origin of life on Earth.

DATA AVAILABILITY
The data underlying this article were derived from sources in the public domain and are available in CASSIS, the Combined Atlas of Sources with Spitzer IRS Spectra, at https://cassis.sirtf.com/.

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Figure 1.
The Perseus Molecular Cloud as observed by NASA´s Spitzer Space Telescope showing the location of the IC 348 star cluster and encircled the region containing the IC 348 pointings listed in Table 1. NASA/JPL-Caltech - https://photojournal.jpl.nasa.gov/figures/PIA23405_fig2.jpg
Figure 2. CASSIS spectra of RNO 90 provided by the full aperture and optimal differential extraction techniques.
Figure 3. The three most intense H$_2$ transitions in the 10-30 µm region (S(0), S(1), S(2)) detected in the combined spectrum of the ISM in the IC 348 star-forming region. The central wavelengths provided by the fits show consistency within 0.01 µm with the laboratory wavelengths of these bands.
Figure 4. Excitation diagram of H$_2$ in the gas of the IC 348 star-forming region. The solid black line shows the single-temperature model fit to the three measurements and the blue and red dashed lines are two-temperature fits (each using only two of the measurements).
Figure 5a. HITRAN synthetic absorption spectra (240K and 0.001 atm) of several diatomic molecules and atomic hydrogen in the 10-19.5 μm spectral range
Figure 5b. HITRAN synthetic absorption spectra (as in previous figure)
Figure 6. HITRAN synthetic absorption spectra of OH for several temperatures (75, 240, 300 and 500 K) in the spectral range 11.5-32 µm.
Figure 7. Observed spectrum of IC 348 interstellar gas for the spectral range 10.7-12.0 µm with OH transitions marked.
Figure 8. Caption as in Fig. 7 for the spectral range 13.0-14.8 μm.
Figure 9. HITRAN based computations for molecules detected in diverse astrophysical environments: $\text{H}_2\text{O}$, $\text{CO}_2$, HCN, NO$_2$, N$_2$O.
Figure 10a. HITRAN synthetic water spectra at 50, 100, 150 and 240 K.
Figure 10b. Caption as in figure 10a for the spectral range 19.5-31.0 μm
Figure 11. Comparison of the observed IC 348 ISM spectrum (red solid line) with HITRAN spectral synthesis of $\text{H}_2\text{O} (240K, 0.001\text{atm})$. 
Figure 12. The panels show different lines of H$_2$O in the IC 348 spectrum (red line) and HITRAN (240K, 0.001atm) computations (blue line) at resolution close 600.
Figure 13. Excitation diagram for water transitions detected in the ISM spectrum of IC 348. The solid line is a linear least square fit to the data resulting in excitation temperature $T = 380\, \text{K}$.
Figure 14. Comparison of the CO$_2$ synthetic HITRAN spectrum (solid black line) with the observed spectrum of the ISM in IC 348 (red line). The synthetic spectrum is scaled to match the strength of the 14.98 µm emission band. Other emission features that could be significantly contributed by CO$_2$ lines are marked.
Figure 15. Comparison of HCN synthetic HITRAN spectrum with the observed ISM spectrum in IC 348. The synthetic spectrum (black line) is scaled to match the strength of the 14.04 µm emission feature which is ascribed to the HCN v2 ro-vibrational band at 14.043 µm. Other observed emission features that could have a significant contribution from HCN are marked.
Figure 16. Synthetic HITRAN spectra for tetra-atomic molecules considered in this work.
Figure 17. Comparison of C$_2$H$_2$ synthetic (HITRAN) spectrum with ISM observations in IC 348. The synthetic spectrum (black line) is scaled to match the emission feature at 13.71 µm and the strongest ro-vibrational band of C$_2$H$_2$. The emission lines at 13.675 and 13.898 µm could have a significant contribution from C$_2$H$_2$. 
Figure 18. Comparison of NH$_3$ synthetic (HITRAN) bands with the ISM observed IC 348 spectrum. The synthetic spectrum has been scaled to match the strength of the observed 13.74 $\mu$m feature. NH$_3$ may also contribute to the waker emission at 10.34 micron.
Figure 19. HITRAN synthetic spectra for polyatomic molecules at 240 K.
Figure 20. Identification of bands of several polyatomic molecules (CO$_2$, C$_2$H$_2$, C$_2$H$_6$, C$_4$H$_2$, C$_6$H$_2$, C$_6$H$_6$, HCN, HC$_3$N, HC$_5$N) in the spectrum of the ISM in IC 348.
Figure 21. Fit to the emission feature ascribed to benzene $v_4$ Q-branch at 14.837 µm.
Figure 22. Tryptophan (Trp) bands: laboratory wavelengths are marked with vertical lines. Emission features assigned to these bands are indicated (yellow filled) in the “combined IC 348 ISM” spectrum (red colour) and in the “combined star-forming region ISM” spectrum (black). Blue asterisks mark the position of water bands. The spectra are shifted on the vertical axis for convenience of display, only band fluxes relative to the local continuum can be deduced from these plots.
Figure 23. Tyrosine (Tyr) bands: caption as in Fig. 22.
Figure 24. Phenylalanine (Phe) bands: caption as in Fig. 22.
Figure 25. Isoleucine (Ile) bands: caption as in Fig. 22.
Figure 26. Glycine (Gly) bands: caption as in Fig.22
Figure 27. Vibrational excitation diagram for the bands of three aromatic amino acids, isoleucine and glycine observed in the ISM of IC 348 (Perseus Molecular Cloud). Natural logarithm of $N_u/g_u$ versus energy $E_u/k$ of the excited vibrational states for all the observed amino acid bands with available fluxes and integrated molar absorptivities. Tryptophan transitions (black triangles), tyrosine (blue diamond), phenylalanine (green crosses), isoleucine (violet squares) and glycine (red circles).
Figure 28. UV absorbance of amino acids. Molar extinction coefficients as a function of wavelength (Wetlaufer 1963).
Table 1. IC 348 ISM locations observed with Spitzer IRS

Short-High spectra

| AOR   | R.A._2000    | Dec._2000   |
|-------|--------------|-------------|
| 22848000 c | 03:44:38.90  | +32:10:03.0 |
| 22848512 c | 03:44:41.66  | +32:06:45.4 |
| 22849024 c | 03:45:00.87  | +32:09:15.5 |
| 22849536 c | 03:44:22.88  | +32:04:57.0 |
| 22850048 c | 03:44:42.71  | +32:03:29.8 |
| 22850560 c | 03:44:36.08  | +32:00:14.7 |
| 22851072 c | 03:43:50.10  | +32:08:17.9 |
| 22851584 c | 03:44:33.23  | +31:59:54.1 |

Long-High spectra

| AOR   | R.A._2000    | Dec._2000   |
|-------|--------------|-------------|
| 27542016_4 w | 03:44:32.67  | +32:11:05.2 |
| 27542016_2 w | 03:44:30.31  | +32:11:35.2 |
| 27542016_3 w | 03:44:27.95  | +32:12:05.2 |
| 27542016_1 w | 03:44:30.31  | +32:11:35.2 |

^c program ID 40247 (N. Calvet, P.I.)
^w program ID 50560 (D. Watson, P.I.)
Table 2. Spitzer spectroscopic observations of various ISM locations

| AOR           | R.A. 2000 | Dec. 2000 |
|---------------|-----------|-----------|
| 27058432      | 04:33:43.51 | +25:20:38.1 |
| 24404480      | 04:35:27.37 | +24:15:58.9 |
| 27062784      | 16:26:23.42 | -24:20:00.5 |
| 27057152      | 04:42:41.01 | +25:15:37.3 |
| 27067136      | 04:47:11.45 | +16:58:42.9 |
| 27059712      | 18:30:04.90 | +01:15:06.5 |
| 27063808      | 16:02:57.80 | -40:18:25.3 |
| 27062528      | 04:14:14.59 | +28:28:43.0 |
| 27060224      | 04:23:42.12 | +24:56:14.3 |
| 27058688      | 04:23:39.18 | +24:56:14.3 |
| 27060224      | 04:23:42.12 | +24:56:14.3 |
| 27062016      | 16:31:33.46 | -24:28:37.4 |
| 27059200      | 04:31:53.51 | +24:24:17.8 |
| 27059200      | 04:31:53.51 | +24:24:17.8 |
| 27057664      | 04:35:50.22 | +22:49:41.9 |
| 27064832      | 15:45:08.02 | -34:18:15.8 |
| 27064320      | 15:56:03.25 | -37:56:06.3 |
| 27063552      | 03:30:44.01 | +30:33:47.0 |
| 27061760      | 16:34:05.01 | -15:48:16.8 |
| 27064064      | 15:56:42.31 | -37:49:15.4 |
| 27066112      | 11:08:15.49 | -77:41:43.6 |
| 27065856      | 11:09:36.51 | -76:23:20.7 |
| 27066112      | 11:08:15.49 | -77:41:43.6 |
| 24402944      | 11:01:55.52 | -34:43:02.1 |
| 27066880      | 04:56:04.03 | -30:34:01.3 |
| 27066624      | 10:55:59.76 | -77:23:55.1 |
| 27066368      | 10:59:10.00 | -77:23:10.0 |
| 27065088      | 13:00:41.87 | -77:10:22.1 |
| 24402944      | 11:01:55.52 | -34:43:02.1 |
| 27060736      | 16:48:42.53 | -14:15:51.0 |
| 27065600      | 11:10:12.73 | -77:37:09.0 |
| 27065344      | 11:11:39.66 | -76:21:00.3 |

`c` program ID 50641,20363 (J. Carr, PI)

`n` program ID 30300 (J. Najita, PI)
Table 3. Measured fluxes of water lines in the spectrum of RNO 90
(ID program: 50641; AOR 27061760)

| Wavelength (µm) | Flux\textsuperscript{b} \((10^{17} \text{ W m}^{-2})\) | Flux\textsuperscript{c} \((10^{17} \text{ W m}^{-2})\) |
|-----------------|-----------------|-----------------|
| 15.16 + 15.17   | < 5.8           | 5.7             |
| 15.57 + 15.62   | 18.0 ± 1.0      | 16.5            |
| 15.74           | 7.3 ± 0.6       | 7.2             |
| 17.10           | 9.0 ± 0.9       | 9.5             |
| 17.23           | < 9.4           | 9.2             |
| 22.54+22.62+22.64 | 12.5 ± 1.0   | 14.0            |
| 23.46 + 23.51   | 12.8            | 13.0            |
| 26.42           | < 2.0           | 2.2             |
| 28.59           | 11.5 ± 0.6      | 9.0             |
| 29.14           | < 4.6           | 3.9             |
| 29.36           | < 0.8           | <1.0            |
| 30.47 +30.53    | 12.2 ± 0.7      | 13.5            |
| 30.87 +30.90    | 10.5 ± 0.8      | 10.5            |
| 31.74 +31.77    | < 2.6           | 2.5             |
| 32.80 +30.83    | < 3.2\textsuperscript{b} | 2.7             |
| 32.92 +32.99+33.01 | 27.4 ± 4.9   | 25.0            |

\textsuperscript{b}Blevins et al (2016)
\textsuperscript{c} This work (full aperture)
### Table 4. H$_2$ in IC 348 gas

| Transition | $\lambda$ [μm] | $E_u$ [K] | $A_{ul}$ [s$^{-1}$] | $g_u$ | $F$ ($10^{-18}$ Wm$^{-2}$) | $N_u$ (cm$^{-2}$) |
|------------|----------------|-----------|----------------------|------|-----------------------------|------------------|
| H$_2$ v = 0–0 S(0) J = 2–0 | 28.219 | 509.9 | 2.943($-11$) | 5 | 25± 2 | 2.6E+20 |
| H$_2$ v = 0–0 S(1) J = 3–1 | 17.035 | 1015.1 | 4.761($-10$) | 21 | 43±4 | 7.8E+19 |
| H$_2$ v = 0–0 S(2) J = 4–2 | 12.279 | 1681.9 | 2.755($-9$) | 9 | 22±2 | 5.8E+18 |

Note: molecular parameters from Roueff et al. (2019) and HITRAN database
### Table 5. H$_2$O in the gas of IC 348

| Transit. | Wavelength (μm) | Wavenum (cm$^{-1}$) | $A_{ij}$ (s$^{-1}$) | $E_{up}$ (K) | $g$ | $F_{obs}$ (10$^{-16}$ Wm$^{-2}$) | $N_{ij}$ (10$^{13}$ cm$^{-2}$) | Notes |
|----------|-----------------|----------------------|-------------------|-------------|-----|-------------------|-----------------|-------|
| o-H$_2$O$^{a}$ 16.41 - 15.15 | 12.375 | 806.696 | 7.61 | 4948 | 105 | <1 | -- | b |
| p-H$_2$O$^{a}$ 11.3 - 10.0 | 12.407 | 805.996 | 4.19 | 4945 | 33 | <1 | -- | u |
| o-H$_2$O$^{a}$ 11.8 - 10.5 | 12.444 | 803.546 | 0.29 | 3629 | 69 | 20±2 | 0.43 | str+blend |
| o-H$_2$O$^{a}$ 13.7 - 12.4 | 12.453 | 802.989 | 1.04 | 4213 | 69 | <1 | -- | u |
| o-H$_2$O$^{a}$ 10.2 - 9.3 | 12.683 | 779.303 | 0.15 | 3217 | 21 | <1 | -- | u |
| o-H$_2$O$^{a}$ 12.1 - 11.0 | 15.738 | 635.397 | 1.10 | 2823 | 75 | <1 | -- | u |
| o-H$_2$O$^{a}$ 12.5 - 11.2 | 17.103 | 584.708 | 3.78 | 3273 | 75 | 5±1 | 0.114 | pr |
| p-H$_2$O$^{a}$ 11.3 - 10.0 | 17.225 | 580.536 | 0.97 | 2438 | 23 | 5±1 | 0.446 | r |
| o-H$_2$O$^{a}$ 11.2 - 10.1 | 17.358 | 576.114 | 0.96 | 2432 | 69 | 5±1 | 0.461 | r |
| o-H$_2$O$^{a}$ 10.8 - 9.7 | 22.538 | 443.695 | 33.15 | 3243 | 63 | 3±1 | 0.002 | b |
| o - H$_2$O$^{a}$ 6.62 - 5.63 | 22.619 | 442.088 | 0.06 | 1278 | 39 | 2±1 | 0.82 | b |
| o-H$_2$O$^{a}$ 5.20 - 4.21 | 22.639 | 441.714 | 0.014 | 1067 | 33 | 4±1 | 7.04 | s |
| p-H$_2$O$^{a}$ 5.21 - 4.22 | 23.458 | 426.293 | 0.015 | 1067 | 11 | 13±2 | 22.15 | vs |
| p-H$_2$O$^{a}$ 7.71 - 6.40 | 28.591 | 349.755 | 20.39 | 2006 | 15 | 4±1 | 0.006 | sr |
| p-H$_2$O$^{a}$ 9.55 - 8.44 | 29.137 | 343.205 | 8.98 | 2122 | 19 | 2±0.5 | 0.007 | r |
| o-H$_2$O$^{a}$ 7.25 - 6.16 | 29.836 | 335.157 | 0.51 | 1100 | 45 | 3±1 | 0.19 | r |
| o-H$_2$O$^{a}$ 7.61 - 6.55 | 30.525 | 327.595 | 13.58 | 1749 | 45 | <1 | -- | u |
| o-H$_2$O$^{a}$ 7.62 - 6.44 | 30.529 | 327.557 | 13.57 | 1735 | 15 | <1 | -- | u |

$^{a}$Blevins et al 2019; $^{b}$HiTRAN ; s-strong, sr-strong resolved, vs-very strong, b-blend, pr-partially resolved, r-resolved, u undetected
Table 6. Laboratory wavelengths and integrated absorptivities, $\psi$, for amino acid mid-IR transitions. Wavelength and flux measurements for bands in the IC 348 combined (averaged) ISM spectrum

### Tryptophan

| Wavenumber_{lab} (cm$^{-1}$) | Wavelength_{lab} (µm) | Wavelength_{obs} (µm) | $\psi$ (km mol$^{-1}$) | Flux ($10^{-18}$ W m$^2$) |
|-------------------------------|------------------------|------------------------|-------------------------|-----------------------------|
| 920                           | 10.87                  | 10.87                  | 4.4                     | 5.6                         |
| 865                           | 11.56                  | 11.58                  | 10.8                    | 4.5                         |
| 744                           | 13.43                  | 13.45                  | 30.6                    | 6.4                         |
| 683                           | 14.65                  | 14.66                  | 2.3                     | 1.4 blend Phe               |
| 627                           | 15.96                  | 15.96                  | 2.7                     | 1.5                         |
| 588                           | 17.00                  | 16.98                  | --                      | <1.0                        |
| 580                           | 17.23                  | 17.23                  | 5.6                     | 4.5 blend $H_2O$            |
| 559                           | 17.88                  | 17.88                  | 9.8                     | 2.8                         |
| 550                           | 18.19                  | 18.23                  | --                      | 2.6                         |
| 528                           | 18.92                  | 18.93                  | --                      | 1.5                         |
| 509                           | 19.64                  | 19.63                  | 31.7                    | 3.0                         |
| 499                           | 20.02                  | 20.02                  | --                      | 1.5                         |
| 457                           | 21.88                  | 21.88                  | 1.9                     | 2.0                         |
| 426                           | 23.46                  | 23.47                  | 10.0                    | 4.8 blend $H_2O$            |
| 357                           | 28.03                  | 28.02                  | --                      | 1.5                         |
| 349                           | 28.65                  | 28.70                  | 9.6                     | 3.0                         |
| 324                           | 30.85                  | 30.75                  | 3.2                     | <3.0                        |

### Tyrosine

| Wavenumber_{lab} (cm$^{-1}$) | Wavelength_{lab} (µm) | Wavelength_{obs} (µm) | $\psi$ (km mol$^{-1}$) | Flux ($10^{-18}$ W m$^2$) |
|-------------------------------|------------------------|------------------------|-------------------------|-----------------------------|
| 985                           | 10.15                  | 10.14                  | 2.8                     | 1.0                         |
| 897                           | 11.15                  | 11.15                  | 2.3                     | <1.2                        |
| 877                           | 11.40                  | 11.45                  | 4.6                     | 5.0                         |
| 841                           | 11.89                  | 11.90                  | 14.5                    | 3.7                         |
| 794                           | 12.59                  | 12.60                  | 12.7                    | 3.0                         |
| 740                           | 13.50                  | 13.51                  | 4.9                     | 2.5                         |
| 650                           | 15.39                  | 15.40                  | 12.7                    | 2.2                         |
| 576                           | 17.36                  | 17.34                  | 12.7                    | 6.8 blend $H_2O$+Full      |
| 530                           | 18.86                  | 18.89                  | 16.0                    | 12.0 blend Full             |
| 494                           | 20.26                  | 20.25                  | 2.7                     | 2.9                         |
| 434                           | 23.05                  | 23.01                  | 5.5                     | 3.0                         |
| 380                           | 26.32                  | 26.34                  | 15.6                    | 3.4                         |
| 335                           | 29.80                  | 29.72                  | 1.3                     | 2.2                         |
### Phenylalanine

| bp  | Value1 | Value2 | Value3 | Value4 | Note          |
|-----|--------|--------|--------|--------|---------------|
| 950 | 10.52  | 10.52  | 7.2    | 5.7    | blend $C_2H_4$|
| 914 | 10.94  | 10.95  | 1.7    | 2.4    |               |
| 779 | 12.84  | 12.86  | 3.6    | 3.0    |               |
| 746 | 13.38  | 13.39  | 9.2    | <1.5   |               |
| 700 | 14.29  | 14.29  | 19.8   | 6.9    |               |
| 526 | 18.98  | 18.98  | 4.0    | 7.2    |               |
| 469 | 21.34  | 21.35  | 5.1    | 3.0    |               |
| 366 | 27.31  | 27.33  | 54.0   | 10.0   |               |

### Isoleucine

| bp  | Value1 | Value2 | Value3 | Value4 | Note          |
|-----|--------|--------|--------|--------|---------------|
| 921 | 10.85  | 10.84  | 3.0    | <1.0   |               |
| 873 | 11.45  | 11.46  | 3.2    | 2.5    |               |
| 712 | 14.05  | 14.05  | 5.4    | 2.7    | blend $HCN$   |
| 557 | 17.94  | 17.95  | ---    | 3.5    |               |
| 538 | 18.59  | 18.58  | 12.9   | 5.0    |               |
| 443 | 22.55  | 22.56  | 7.0    | 3.0    | blend $H_2O$  |
| 393 | 25.42  | 25.38  | 5.2    | <1.4   |               |
| 343 | 29.13  | 29.14  | 4.5    | 3.4    | blend $H_2O$  |

### Glycine

| bp  | Value1 | Value2 | Value3 | Value4 | Note          |
|-----|--------|--------|--------|--------|---------------|
| 910 | 10.99  | 10.98  | ---    | 4.0    |               |
| 893 | 11.20  | 11.19  | 10.5   | < 4.0  | blend $PAH$   |
| 698 | 14.32  | 14.33  | 6.6    | 13.0   |               |
| 608 | 16.45  | 16.45  | 2.1    | 6.0    |               |
| 504 | 19.85  | 19.86  | 15.1   | 4.5    |               |
| 359 | 27.85  | 27.75  | 14.4   | 3.4    |               |

*bp* Blend partially
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