

## GC – MS Determination of Bioactive Components of *Rorippa indica* L.

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**Abstract:** *Rorippa indica* L. is one of the medicinally plant belonging to the family Brassicaceae, commonly known as water cress. In present study the methanol extract of seeds and roots of *Rorippa indica* have been subjected to GC-MS analysis, twenty five phytochemical compounds are identified. The identification is performed by the index of NIST and WILEY libraries.

**Key words:** GC-MS analysis, Bioactive Components, *Rorippa indica*, Methanol Extract.

### Introduction

*Rorippa indica* L. belongs to Brassicaceae family commonly known as water cress. It is a wild crucifer found in all continents except Antarctica, mainly with dense population throughout India. It is distributed in plains of Coimbatore, North Arcot and Tiruchirappalli districts of Tamil Nadu<sup>1</sup>. It is aphid tolerant and contains high percentage of oleic acid (26.65%), linoleic acid (26.94%) and a moderate percentage of crucic acid (24.40%)<sup>2</sup>. Basal leaves often lyrate or pinnatifid, upper ones simple 6-12x 3-4 cm, chartaceous, base attenuate, margin sinuate or irregularly toothed, apex obtuse, petiole much reduced, aplexicaul. Racemes terminal and or leaf opposed to 15 cm pedicel to 2 mm. Flowers 4 mm across. Sepal 4, elliptic, 2 mm concave, not saccate at base. Petals 4, to 3 mm clawed. Disc-nectories 6. Stamens 6; filaments 1.8 and 2 mm; anthers ovoid. Ovary elongate, subsessile, to 2 mm; ovules ; style to 0.3 mm; stigma obscurely 2-lobed or entire. *R. indica* is well known for its medicinal properties such as antiscorbutic, depurative, diuretic, expectorant, hypoglycemic, odontalgic, purgative and stimulants<sup>3</sup>. Taking into consideration of the medicinal importance of this plant, the methanolic extract of seeds and roots of *R.indica* were analyzed for the first time using GC- MS. This work will help to identify the compounds of therapeutic value. GC-MS is one of the best techniques to identify the bioactive constituents of ketone, alcohols, acids, and ester.

### Materials and Methods

#### **Extraction of plant samples**

Seeds and roots of *Rorippa indica* were collected from the hardened regenerated plants. Department of Plant Science Bharathidasan University, Tiruchirappalli, Tamil Nadu. Seeds and roots were cleaned, shade dried and pulverized to powder by a mechanical grinder. Required quantity of powder was weighed and transferred to Soxhlet extraction process and treated with methanol until the powder is fully immersed. The extract was collected and evaporated to dryness by using vacuum distillation unit. The final residue thus obtained was then

analyzed by GC-MS analysis. The spectra of the compounds were matched with NIST and Wiley library. Their structures were defined by the % similarity values.

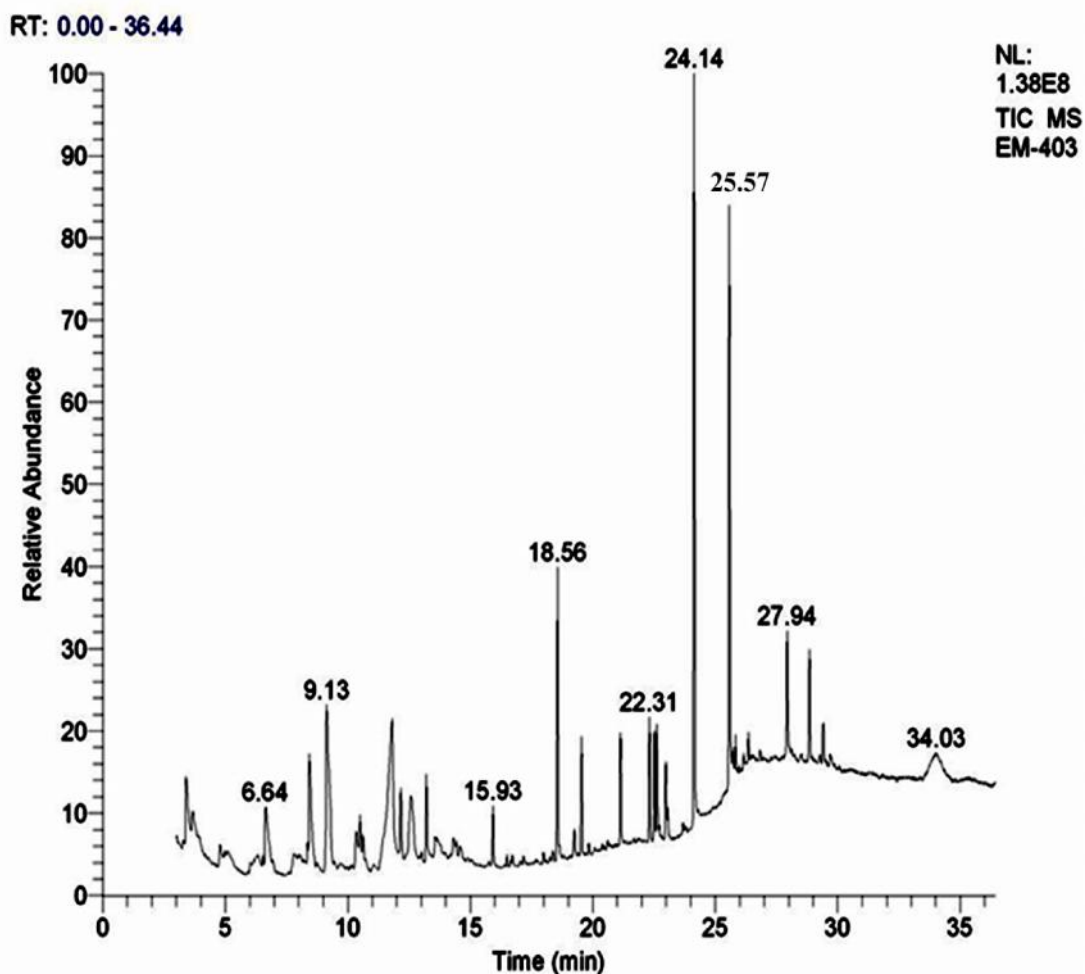
### GC-MS analysis

GC-MS analysis of these extracts were performed using a Thermo GC- Trace ultra version 5.0 and Thermo MS DSQ 11, Colum TR 5- MS- Capillary non polar column (30 Mts, ID : 0.25 mm, FILM : 0.25  $\mu$ m). For GC- MS detection, an electron ionization system with ionizing energy of 70 eV was used. Helium gas was used as the carrier gas at constant flow rate (He, flow: 1 ML/Min), injector temperature 100 - 250, rate: 8/Min, holding time: 10 min @ 250. Total GC running time was 33.43 minutes. The relative % amount of each component was calculated by comparing its average peak area to the total areas, software adopted to handle mass spectra and chromatograms was a turbomass.

**Table 1: Phytocompounds in seed of *Rorippa indica* L. (Methanolic extract)**

S.No	*RT	Name of the Compounds	Molecular Formula	MW	Peak Area %
1	3.42	1-Decene-3,4-dione	C10H16O	168	2.34
2	5.12	2-Iodo-1-methoxy-1-methylcyclohexane	C8H15IO	254	0.98
3	6.66	N,N-Diallylcyanamide	C7H10N2	122	2.22
4	8.44	1,13-Tetradecadiene (CAS)	C14H26	194	4.40
5	9.15	2-Methoxy-4-vinylphenol	C9H10O2	150	7.77
6	10.50	3-(2-Oxocyclopentyl)-2,4-pentanedione	C10H14O3	182	4.45
7	11.79	3-Butyl-1,1-difluoro-2,2-dimethylcyclopropane	C9H16F2	162	6.63
8	12.16	5-Hexyl-2-pentylpyrrolidine	C15H31N	225	1.74
9	12.59	1,6-Anhydro-Beta-D-Glucopyranose (Levogluconan)	C6H10O5	162	4.09
10	13.21	1-Nitro-2,2,3-triphenylpropane	C21H19NO2	317	2.04
11	13.61	Octadecanoic acid (CAS)	C18H36O2	284	1.31
12	14.33	n-Hexdecanioc acid ethyl ester	C18H36O2	284	0.99
13	15.93	Methyl 3-methoxy-4-phenylbutanoate	C12H16O3	208	1.28
14	18.56	Propenoic acid, methyl ester	C11H12O4	208	5.89
15	19.54	Octadecanoic acid, methyl ester (CAS)	C19H38O2	298	2.80
16	21.13	4-methyl-4-nitropentanitrile	C6H10N2O2	142	2.48
17	22.31	Theaspirane A	C13H22O	194	2.79
18	22.62	(+/-)-trans-2-(2,5-Octadiynyl)-3-undecyloxirane	C21H34O	302	4.54
19	22.99	Ferrocenecarboxaldehyde	C13H10FeO	238	2.23
20	24.14	<b>1-Chloro-1-methylcyclohexane</b>	C7H13Cl	132	<b>17.34</b>
21	25.57	5-{Ethoxycarbonyl}-N(1)-[2'-(N",N"-dimethylamino)ethyl]-pyrimido[1,2-a]benzimidazol-4(10H)-one	C17H20N4O3	328	11.38
22	27.94	6-Iodohexanoic acid	C6H11IO2	242	3.12
23	28.85	(+/-)-trans-2-(2,5-Octadiynyl)-3-undecyloxirane	C21H34O	302	2.80
24	29.42	(Z)-(2S,3S)-2,3-bis[(methoxymethyl)oxy]-5-(4-methoxyphenyl)pent-4-enol	C16H24O6	312	1.30
25	34.03	2-Octyldodecan-1-ol	C20H42O	298	3.10

\*RT: Retention Time

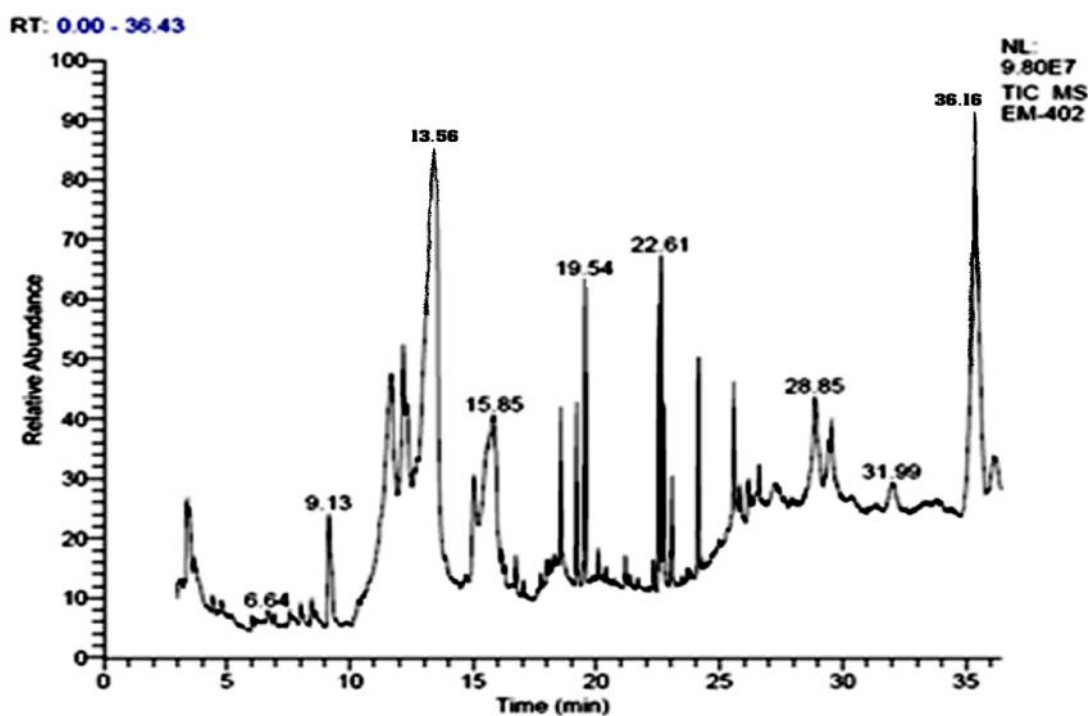
Figure 1: GC-MS chromatograph of methanolic seed extract of *Rorippa indica* L.Table 2: Phytochemicals in root of *Rorippa indica* L. (Methanolic extract)

S.No	*RT	Name of the compounds	Molecular Formula	MW	Peak Area %
1	3.40	7,7,8,8,9,9,10,10,10-Nonadeuteriodec-5-enyl acetate	C <sub>12</sub> H <sub>13</sub> D <sub>9</sub> O <sub>2</sub>	198	3.01
2	8.46	Butyl cis-2-(Cyanomethyl)cyclobutanecarboxylate	C <sub>11</sub> H <sub>17</sub> NO <sub>2</sub>	195	0.82
3	9.15	2-Methoxy-5-vinylphenol	C <sub>9</sub> H <sub>10</sub> O <sub>2</sub>	150	3.78
4	11.67	(+)-1D-cyclohex-5-ene-1,2,4/3-tetrol	C <sub>6</sub> H <sub>10</sub> O <sub>4</sub>	146	7.18
5	12.16	(-)-Conduritol F	C <sub>6</sub> H <sub>10</sub> O <sub>4</sub>	146	5.13
6	13.21	(3S,5S)-5-Ethyl-3,5-dihydroxy-oct-7-en-4-one	C <sub>10</sub> H <sub>18</sub> O <sub>3</sub>	186	1.73
7	13.51	(3S,5S)-5-Ethyl-3,5-dihydroxy-oct-7-en-4-one	C <sub>10</sub> H <sub>18</sub> O <sub>3</sub>	186	13.56
8	15.02	Octadecanoic acid (CAS)	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	2.77
9	15.51	Neopentyl 2-hydroxy-3,3-dimethylbutanoate	C <sub>11</sub> H <sub>22</sub> O <sub>3</sub>	202	0.77
10	15.85	Neopentyl 2-hydroxy-3,3-dimethylbutanoate	C <sub>11</sub> H <sub>22</sub> O <sub>3</sub>	202	4.56
11	18.56	(-)-(1S,10bR)-1-Hydroxy-1,2,3,5,6,10b-hexahydropyrrolo[2,1-a]isoquinoline	C <sub>12</sub> H <sub>15</sub> NO	189	2.40

12	19.20	13-Octadecenoic acid, methyl ester (CAS)	C19H36O2	296	2.72
13	19.54	Octadecanoic acid, methyl ester (CAS)	C19H38O2	298	5.42
14	22.61	9-Octadecenoic acid (Z)-, ethyl ester (CAS)	C20H38O2	310	11.58
15	23.06	Tricosanoic acid, methyl ester (CAS)	C24H48O2	368	2.15
16	24.14	1-Chloro-1-methylcyclohexane	C7H13Cl	132	3.53
17	25.58	5-{Ethoxycarbonyl}-N(1)-[2'-(N",N"-dimethylamino)ethyl]-pyrimido[1,2-a]benzimidazol-4(10H)-one	C17H20N4O3	328	2.11
18	26.16	Heneicosanoic acid, methyl ester (CAS)	C22H44O2	340	1.08
19	26.59	N-(tert-Butoxycarbonyl)-2-methyl-4,4-diphenylbut-3-enamine	C22H27NO2	337	0.91
20	27.26	[2-[(phenylsulfonyl)methyl]allyl]trimethylsilane	C13H20O2	268	1.09
21	28.87	Ethyl 4-Benzyloxy-2-[2-methyl-2(E)-butenyl]-2-[2(E),4-pentadienyl]acetoacetate	C23H30O4	370	2.70
22	29.53	N-(tert-Butoxycarbonyl)-2-methyl-4,4-diphenylbut-3-enamine	C22H27NO2	337	2.71
23	32.02	1-(2-Hydroxyethoxy) tridecane	C15H32O2	244	1.26
24	35.39	(E,E)-1,14-Dibromo-2,12-tetradacadiene	C14H24Br2	350	2.23
25	36.16	<b>Ethyl 4-Benzyloxy-2-[2-methyl-2(E)-butenyl]-2-[2(E),4-pentadienyl]acetoacetate</b>	C23H30O4	370	<b>14.80</b>

\*RT: Retention Time

Figure 2: GC-MS chromatograph of methanolic root extract of *Rorippa indica* L.



**Table 3:** Activity of phytochemicals identified in the Methanol extracts of the seeds of *Rorippa indica* L. by GC-MS

S.No	Name of the Compounds	Compound Nature	*Bioactivity
1	1-Decene-3,4-dione	Ketone Compound	Toxicity
2	2-Iodo-1-methoxy-1-methylcyclohexane	Alcoholic Compound	No report
3	N,N-Diallylcyanamide	Cyanide Compound	Pest control
4	1,13-Tetradecadiene (CAS)		
5	2-Methoxy-4-vinylphenol	Phenolic Compound	Perfumery, Insecticide and Fungicide
6	3-(2-Oxocyclopentyl)-2,4-pentanedione	Ketone Compound	Toxicity
7	3-Butyl-1,1-difluoro-2,2-dimethylcyclopropane	Fluoro Compound	Fluoropolymers, Refrigerants, Solvents, Fluorosurfactants
8	5-Hexyl-2-pentylpyrrolidine	-	No report
9	1,6-Anhydro-Beta-D-Glucopyranose (Levogluconan)	-	No report
10	1-Nitro-2,2,3-triphenylpropane	Nitrogen Compound	Antioxidant
11	Octadecanoic acid (CAS)	Stearic acid	Cancer preventive Insectifuge
12	n-Hexadecanoic acid ethyl ester	Ester Compound	Antioxidant, Nematicide, Pesticide, Flavor, Antiandrogenic
13	Methyl 3-methoxy-4-phenylbutanoate	-	No report
14	Propionic acid, methyl ester	Ester Compound	Flavor, Fungicide, Irritant, Perfumery, Pesticide
15	Octadecanoic acid, methyl ester (CAS)	-	No report
16	4-methyl-4-nitropentanitrile	Nitrogen Compound	No report
17	Theaspirane A	-	No report
18	(+)-trans-2-(2,5-Octadiynyl)-3-undecyloxirane	-	No report
19	Ferrocenecarboxaldehyde	Aldehyde Compound	No report
20	1-Chloro-1-methylcyclohexane	-	No report
21	5-{Ethoxycarbonyl}-N(1)-[2'-(N",N"-dimethylamino)ethyl]-pyrimido[1,2-a]benzimidazol-4(10H)-one	Alcoholic Compound	No report
22	6-Iodoheptanoic acid	-	No report
23	(+)-trans-2-(2,5-Octadiynyl)-3-undecyloxirane	-	No report
24	(Z)-(2S,3S)-2,3-bis[(methoxymethyl)oxy]-5-(4-methoxyphenyl)pent-4-enol	-	No report
25	2-octyldodecan-1-ol	Alcoholic Compound	No report

\*Duke's Phytochemical and Ethnobotanical Databases (online Database)

**Table 4:** Activity of phyto- components identified in the Methanol extracts of the roots of *Rorippa indica* by GC-MS.

S.No	Name of the compounds	Compound Nature	*Biological activity
1	7,7,8,8,9,9,10,10,10-Nonadeuteriodec-5-enyl acetate	-	No report
2	Butyl cis-2-(Cyanomethyl)cyclobutanecarboxylate	Cyanide	Pest control
3	2-Methoxy-5-vinylphenol	Phenolic Compound	Perfumery, Insecticide and Fungicide
4	(+)-1D-cyclohex-5-ene-1,2,4/3-tetrol	-	No report
5	(-)-Conduritol F	-	No report
6	(3S,5S)-5-Ethyl-3,5-dihydroxy-oct-7-en-4-one	Ketone Compound	Toxicity
7	(3S,5S)-5-Ethyl-3,5-dihydroxy-oct-7-en-4-one	Ketone Compound	Toxicity
8	Octadecanoic acid (CAS)	Stearic acid	Cancer preventive Insectifuge
9	Neopentyl 2-hydroxy-3,3-dimethylbutanoate	-	No report
10	Neopentyl 2-hydroxy-3,3-dimethylbutanoate	-	No report
11	(-)-(1S,10bR)-1-Hydroxy-1,2,3,5,6,10b-hexahydropyrrolo[2,1-a]isoquinoline	-	No report
12	13-Octadecenoic acid, methyl ester (CAS)	Ester Compound	Antiinflammatory
13	Octadecanoic acid, methyl ester (CAS)	Ester Compound	No report
14	9-Octadecenoic acid (Z)-, ethyl ester (CAS)	Ester Compound	No report
15	Tricosanoic acid, methyl ester (CAS)	Ester Compound	No report
16	1-Chloro-1-methylcyclohexane	-	No report
17	5-{Ethoxycarbonyl}-N(1)-[2'-(N",N"-dimethylamino)ethyl]-pyrimido[1,2-a]benzimidazol-4(10H)-one	-	No report
18	Heneicosanoic acid, methyl ester (CAS)	--	No report
19	N-(tert-Butoxycarbonyl)-2-methyl-4,4-diphenylbut-3-enamine	-	No report
20	[2-[(phenylsulfonyl)methyl]allyl]trimethylsilane	-	No report
21	Ethyl 4-Benzyloxy-2-[2-methyl-2(E)-butenyl]-2-[2(E),4-pentadienyl]acetoacetate	-	No report
22	N-(tert-Butoxycarbonyl)-2-methyl-4,4-diphenylbut-3-enamine	--	No report
23	1-(2-Hydroxyethoxy)tridecane	---	No report
24	(E,E)-1,14-Dibromo-2,12-tetradacadiene	--	No report
25	Ethyl 4-Benzyloxy-2-[2-methyl-2(E)-butenyl]-2-[2(E),4-pentadienyl]acetoacetate	-	No report

\* Dr.Duke's Phytochemical and Ethnobotanical Databases (online Database)

## Results and Discussion

The components present in the methanolic extracts of seeds and roots of *R. indica* were identified by GC-MS analysis (Figure 1 and 2). The active principles with their Retention Time (RT), Molecular formula, Molecular Weight (MW) and Concentration (%) in the seeds and roots of *R. indica* are presented in Table (1 and 2).

Twenty five compounds were identified in the methanol seed extract of *R. indica*. The results revealed that 1-Chloro-1-methylcyclohexane (17.34%), was found as major compound followed by 5-{Ethoxycarbonyl}-N(1)-[2'-(N",N"-dimethylamino)ethyl]-pyrimido [1,2-a] benzimidazol-4(10H)-one (11.38%), 2-Methoxy-4-

vinylphenol (7.77%) and 3-Butyl-1,1-difluoro-2,2-dimethylcyclopropane (6.63%) (Table1). Similarly twenty five compounds were identified in the methanol root extracts of *R. indica*. Major compound in the root extract are Ethyl 4-Benzoyloxy-2-[2-methyl-2(E)-butenyl]-2-[2(E),4-pentadienyl] acetate (14.80%), 3S, 5S-5-Ethyl-3, 5-dihydroxy-oct-7-en-4-one (13.56%) and 9-Octadecenoic acid (Z)-, ethyl ester (CAS) (11.58%) (Table 2). 2-Methoxy-5-vinylphenol, Octadecanoic acid (CAS), Octadecanoic acid, methyl ester (CAS) and 1-Chloro-1-methylcyclohexane are distributed in both seed and root of *R. indica*.

The phytochemicals and their biological activities obtained through the GC-MS analysis of *Rorippa indica* seeds and roots were presented in the Table (3 and 4). n- Hexadecanoic acid ethyl ester and Octadecanoic acid have been reported in *Pterocarpus marsupium*<sup>4</sup> and *Calophyllum inophyllum*<sup>5</sup>. The biological activities listed are based on Duke's Phytochemical and Ethnobotanical Database and Jim Duke Agricultural Research Service/ USDA.

Medicinal properties of *R. indica* includes antiscorbutic, depurative, diuretic, expectorant, hypoglycemic, cancer preventive, odontalgic, purgative and stimulants<sup>3</sup>, of these properties, cancer preventive activity is confirmed by the bioactive compound octadecanoic acid from seed as well as root methanolic extracts Table (3 and 4). Besides, *R. indica* also have toxicity properties (1-Decene-3,4-dione and (3S,5S)-5-Ethyl-3,5-dihydroxy-oct-7-en-4-one) from seed and root, pest and insect control quality (N,N-Diallylcyanamide, Butyl cis-2-(Cyanomethyl) cyclobutanecarboxylate, 2-Methoxy-5-vinylphenol, n-hexadecanoic acid ethyl ester, propionic acid, methyl ester) in seed and root Table (3 and 4), antioxidant property (1-Nitro-2,2,3-triphenylpropane, n-hexadecanoic acid ethyl ester) from seed extract anti-inflammatory property (13-Octadecenoic acid, methyl ester) in root<sup>4</sup>.

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