## List of Figures

<table>
<thead>
<tr>
<th>Figure</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Figure 1.1</td>
<td>SEM pictures of <em>Pediococcus acidilactici</em> and <em>Lactobacillus casei</em></td>
<td>2</td>
</tr>
<tr>
<td>Figure 1.2</td>
<td>Photograph showing different natural prebiotic sources</td>
<td>3</td>
</tr>
<tr>
<td>Figure 1.3</td>
<td>Potential applications of prebiotics</td>
<td>5</td>
</tr>
<tr>
<td>Figure 1.4</td>
<td>Combined effect of synbiotics (Probiotics and Prebiotics) on the health of host</td>
<td>5</td>
</tr>
<tr>
<td>Figure 1.5</td>
<td>Various methods of production of prebiotics</td>
<td>9</td>
</tr>
<tr>
<td>Figure 2.1</td>
<td>Chemical structure of inulin compound</td>
<td>25</td>
</tr>
<tr>
<td>Figure 2.2</td>
<td><em>Cichorium intybus</em> (Chicory) a naturally occurring inulin</td>
<td>29</td>
</tr>
<tr>
<td>Figure 2.3</td>
<td>Growth of <em>Pediococcus acidilactici</em> in MRS broth (without glucose) with of 2.0% (MRS-2), 3.0% (MRS-3), 4.0% (MRS-4) and 5.0% (MRS-5) of sugar cane molasses, at initial pH 6.5</td>
<td>38</td>
</tr>
<tr>
<td>Figure 2.4</td>
<td>FT-IR spectrum for inulin from artichoke (a) and chicory (b)</td>
<td>40</td>
</tr>
<tr>
<td>Figure 2.5</td>
<td>GC–MS results for artichoke inulin (a) compared with chicory (b), dahlia, (c) and Jerusalem artichoke (d) inulins</td>
<td>40</td>
</tr>
<tr>
<td>Figure 2.6</td>
<td>TLC analysis of fructose (1) chicory inulin (2) chicory inulin plus inulinase (3) artichoke inulin (4) and artichoke inulin plus inulinase (5)</td>
<td>41</td>
</tr>
<tr>
<td>Figure 2.7</td>
<td>Thin layer chromatography of inulin extraction: Lane 1, direct sonication; Lane 2, indirect sonication; Lane 3, conventional extraction; Lane 4, purified inulin</td>
<td>42</td>
</tr>
<tr>
<td>Figure 2.8</td>
<td>High-performance anion exchange chromatogram of inulins from conventional extraction (a), indirect sonication (b), direct sonication (c)</td>
<td>42</td>
</tr>
<tr>
<td>Figure 2.9</td>
<td>Sugar composition of white-flesh (a) and red-flesh (b) dragon fruit extracts, analysed by HPLC [52]</td>
<td>43</td>
</tr>
</tbody>
</table>
Figure 2.10: UV–vis spectra of silver colloids

Figure 2.11: TEM images of colloid s3 (a) and (b) under different magnification; (c) high-resolution image of single nanocrystal showing lattice fringes with spacing of 0.23nm and (d) SAED pattern

Figure 2.12: TEM images of stevia leaf extract–reduced SNPs. (a) and (b) demonstrate isolated spherical and ellipsoidal nanoparticles of different sizes, while (c) and (d) reveal larger particles with polygonal shapes.

Figure 2.13: UV–vis spectra of the mixture of stevia leaf extract–AgNO3 solution recorded at various reaction times, as shown in the legend

Figure 2.14: FTIR spectra of the stevia leaf extract (light line) and stevia leaf extract–reduced SNPs (dark line)

Figure 2.15: Mechanisms of antibiotic resistance in probiotics. (A) Intrinsic antibiotic resistance a) efflux pumps, b) antibiotic degrading enzyme, c) antibiotic altering enzyme and d) Inner change (B) Acquired antibiotic resistance (a) transformation, (b) conjugation, (c) and transduction

Figure 4.1: Photographs of instruments

Figure 4.2: Process of inulin extraction from natural prebiotic sources

Figure4.3: Photograph of husk free arabinoxylan gel

Figure 4.4: Photograph showing TLC

Figure 5.1.1: a. FESEM image of *Pediococcus acidilactici* in diploid coccal arrangement b. Enlarged view of single cell c. d. *Pediococcus acidilactici* cells treated with prebiotic inulin

Figure 5.1.2: FTIR chromatogram of inulin

Figure 5.1.3: FTIR chromatogram comparison between inulin, garlic, wheat, oat and dalia
Figure 5.1.4: TLC of standard inulin and isolated inulin from different sources 100

Figure 5.1.5: HPLC chromatogram of inulin 102

Figure 5.1.6: HPLC chromatogram of garlic 102

Figure 5.1.7: HPLC chromatogram of oat 103

Figure 5.1.8: HPLC chromatogram of wheat 103

Figure 5.1.9: FTIR of psyllium husk 104

Figure 5.1.10: TLC of standard arabinose, xylose; hydrolysed isolated arabinoxylan; isolated arabinoxylan (crude) 105

Figure 5.1.11: HPTLC of arabinoxylan, arabinose and Xylose 106

Figure 5.2.1: Comparison of predicted and actual values of response variables 110

Figure 5.2.2: Surface plot showing combined effects of Glucose concentration and pH on the biomass concentration of P.acidilactici 110

Figure 5.2.3: Surface plot showing combined effects of inulin concentration and pH on the biomass concentration of Pediococcus acidilactici 110

Figure 5.2.4: Surface plot showing combined effects of Glucose and Inulin concentration on the biomass concentration of Pediococcus acidilactici 111

Figure 5.2.5: Comparison of predicted and actual values of response variables (garlic –inulin) 118

Figure 5.2.6: Comparison of predicted and actual values of response variables (wheat-inulin) 118

Figure 5.2.7: Comparison of predicted and actual values of response variables (oat inulin) 118

Figure 5.2.8: Comparison of predicted and actual values of response variables (dalia inulin) 118
Figure 5.2.9: Surface plot showing combined effects of garlic-inulin concentration and pH on the biomass concentration of *P. acidilactici* 119

Figure 5.2.10: Surface plot showing combined effects of glucose concentration and pH on the biomass concentration of *P. acidilactici* 119

Figure 5.2.11: Surface plot showing combined effects of glucose and garlic-inulin concentration on the biomass concentration of *P. acidilactici* 119

Figure 5.2.12: Surface plot showing combined effects of wheat-inulin concentration and pH on the biomass concentration of *P. acidilactici* 119

Figure 5.2.13: Surface plot showing combined effects of glucose concentration and pH on the biomass concentration of *P. acidilactici* 119

Figure 5.2.14: Surface plot showing combined effects of glucose and wheat-inulin concentration on the biomass concentration of *P. acidilactici* 119

Figure 5.2.15: Surface plot showing combined effects of oat-inulin concentration and pH on the biomass concentration of *P. acidilactici* 120

Figure 5.2.16: Surface plot showing combined effects of glucose and pH on the biomass concentration of *P. acidilactici* 120

Figure 5.2.17: Surface plot showing combined effects of glucose and oat-inulin concentration on the biomass concentration of *P. acidilactici* 120

Figure 5.2.18: Surface plot showing combined effects of dalia-inulin concentration and pH on the biomass concentration of *P. acidilactici* 120
Figure 5.2.19: Surface plot showing combined effects of glucose concentration and pH on the biomass concentration of *P. acidilactici*

Figure 5.2.20: Surface plot showing combined effects of glucose and dalia-inulin concentration and pH on the biomass concentration of *P. acidilactici*

Figure 5.2.21: Cell growth curve of *Pediococcus acidilactici* in absence of inulin

Figure 5.2.22: Double reciprocal plot of Monod equation

Figure 5.2.23: Cell concentration progress curves at different initial inulin concentrations in complete absence of glucose

Figure 5.2.24: Growth curve showing \( \mu \) versus concentration of inulin

Figure 5.2.25: Specific cell growth rate of *Pediococcus acidilactici* as function of inulin concentration with glucose concentration as parameter

Figure 5.2.26: Specific cell growth rate versus isolated inulin concentration plot

Figure 5.2.27: Cell concentration progress curves at different initial glucose concentrations at 10 g/L inulin concentration

Figure 5.2.28: Cell concentration progress curves at different initial glucose concentrations at 20 g/L inulin concentration

Figure 5.2.29: Cell concentration progress curves at different initial glucose concentrations at 30 g/L inulin concentration

Figure 5.2.30: Cell concentration progress curves at different initial glucose concentrations at 40 g/L inulin concentration

Figure 5.2.31: Plot of specific cell growth rate versus extracted inulin concentration with glucose concentration as parameter.

Figure 5.2.4.1: Biomass concentration time history curve with 5 g/L wheat-inulin
Figure 5.2.4.2: Biomass concentration time history curve with 10 g/L wheat-inulin

Figure 5.2.4.3: Biomass concentration time history curve with 15 g/L wheat-inulin

Figure 5.2.4.4: Biomass concentration time history curve with 20 g/L wheat-inulin

Figure 5.2.4.5: Biomass concentration time history curve with 5 g/L oat-inulin

Figure 5.2.4.6: Biomass concentration time history curve with 10 g/L oat-inulin

Figure 5.2.4.7: Biomass concentration time history curve with 15 g/L oat-inulin

Figure 5.2.4.8: Biomass concentration time history curve with 20 g/L oat-inulin

Figure 5.2.4.9: Biomass concentration time history curve with 5 g/L dalia-inulin

Figure 5.2.4.10: Biomass concentration time history curve with 10 g/L dalia-inulin

Figure 5.2.4.11: Biomass concentration time history curve with 15 g/L dalia-inulin

Figure 5.2.4.12: Biomass concentration time history curve with 20 g/L dalia-inulin

Figure 5.2.4.13: Specific cell growth rate as a function of inulin isolated from wheat with glucose as parameter

Figure 5.2.4.14: Specific cell growth rate as a function of inulin isolated from oat with glucose as parameter

Figure 5.2.4.15: Specific cell growth rate as a function of inulin isolated from dalia with glucose as parameter

Figure 5.3.1: Biomass concentration time history curve
Figure 5.3.2: Plot of specific cell growth rate vs. concentration of garlic paste

Figure 5.3.3: Double reciprocal plot of Monod equation for garlic paste system

Figure 5.3.4: Comparison of prebiotic effect of three systems containing inulin at constant initial glucose concentration at 20 g/L

Figure 5.3.5: Cell dynamics plots of biomass concentration against time at different basil leaf extract concentration

Figure 5.3.6: Specific cell growth rate in presence of basil leaf extract

Figure 5.3.7: Double reciprocal plot of $\mu$ vs $C$ of garlic

Figure 5.3.8: Cell dynamics plots of biomass concentration against time at different betel leaf extract concentration

Figure 5.3.9: Double reciprocal plot of specific cell growth rate and betel leaf extract concentrations

Figure 5.4.1: Bar diagram showing prebiotic scores against pathogens

Figure 5.5.1: Photographs showing zone of inhibition

Figure 5.5.2: Progress of zone of inhibition as a function of time

Figure 5.5.3: Plot of zone of inhibition exhibited by Pediococcus acidilactici under synbiotic condition against concentration of garlic

Figure 5.6.1: UV-visible spectrophotometer of basil leaf-silver nanoparticle

Figure 5.6.2: TEM images of basil-silver nanoparticle synthesized in the laboratory

Figure 5.6.3: EDX image of biosynthesised basil-silver nanoparticles

Figure 5.6.4: Comparison of FTIR spectra between basil-leaf extract and basil-AgNps

Figure 5.6.5: XRD image of basil-AgNps
| Figure 5.6.6: | Biomass concentration time history curves with different concentration of basil-AgNPs | 170 |
| Figure 5.6.7: | Double reciprocal plot of specific cell growth rate $\mu$ versus initial basil-AgNPs concentrations | 171 |
| Figure 5.7.1: | UV-visible spectrophotometer of betel leaf- silver nanoparticle | 172 |
| Figure 5.7.2: | TEM images of betel-silver nanoparticle synthesized in the laboratory | 173 |
| Figure 5.7.3: | EDX image of biosynthesised basil-silver nanoparticles | 174 |
| Figure 5.7.4: | Comparison of FTIR spectra between betel-leaf extract and betel-AgNps | 174 |
| Figure 5.7.5: | Biomass concentration time history curves with different concentration of betel-AgNPs | 176 |
| Figure 5.7.6: | Double reciprocal plot of specific cell growth rate $\mu$ versus initial betel-AgNPs concentrations | 177 |
List of Tables

Table 1.1   Sources of probiotics and prebiotics   4
Table 2.1   Top 10 Foods Containing Prebiotics  24
Table 2.2   Inulin-Type Prebiotic Nomenclature  27
Table 2.3   Physico-chemical characteristics of chicory and artichoke inulin  39
Table 2.4   Overview of probiotic LAB with antibiotic resistances  49
Table 2.5   Commercially available probiotic strains in the market  58
Table 4.1  Composition of modified De-Man Rogossa Sharpe Broth medium (Basis: 1L)  80
Table 5.1  Wavelength numbers of prebiotic samples  99
Table 5.2   Rf values of prebiotic food samples and commercially available inulin  100
Table 5.3   Concentrations of inulin in different prebiotic Indian food materials  101
Table 5.4   Percentage of inulin content in the inulin extracts powder  104
Table 5.5   Wavelength numbers of functional groups present in Psyllium husk  105
Table 5.6   Experimental design in term of coded factors and results of Box-Behnken model  108
Table 5.7   Analysis of variance (ANOVA) for the response surface quadratic model (Biomass)  109
Table 5.8   Model summary statistics  109
Table 5.9   Experimental design in term of coded factors and results of Box-Behnken model  112
Table 5.10 Experimental design in term of coded factors and results of Box-Behnken model for inulin isolated from wheat

Table 5.11 Experimental design in term of coded factors and results of Box-Behnken model for inulin isolated from oat

Table 5.12 Experimental design in term of coded factors and results of Box-Behnken model for inulin isolated from dalia

Table 5.13 Analysis of variance (ANOVA) for the response surface quadratic model

Table 5.14 Analysis of variance (ANOVA) for the response surface quadratic model (Biomass) for wheat-inulin

Table 5.15 Analysis of variance (ANOVA) for the response surface quadratic model (Biomass) for oat-inulin

Table 5.16 Analysis of variance (ANOVA) for the response surface quadratic model (Biomass) for dalia-inulin

Table 5.17 Comparison of experimentally observed response variable (specific cell growth rate) with the values predicted from the response equations.

Table 5.18 Experimental values of cell concentrations as a function of time when grown in absence of inulin

Table 5.19 Specific cell concentration as a function of initial glucose concentration

Table 5.20 Cell concentration time histories of *Pediococcus acidilactici* with initial inulin concentration as parameter in complete absence of glucose

Table 5.21 Specific cell growth rate of *Pediococcus acidilactici* at different initial concentration in complete absence of glucose
Table 5.22  Values of intrinsic kinetic parameters present in Equation (8)  129
Table 5.23  Experimental $\mu$ values at different glucose and inulin concentration  130
Table 5.24  Specific cell growth rate of *Pediococcus acidilactici* at different concentration of inulin derived from garlic in complete absence of glucose  132
Table 5.25  Values of intrinsic kinetic parameters present in Equation (8)  133
Table 5.26  Specific cell growth rate of *Pediococcus acidilactici* at different concentration of inulin derived from garlic in complete absence of glucose  133
Table 5.27.1  Biomass concentration time history with initial inulin concentration of 10 g/L  134
Table 5.27.2  Biomass concentration time history with initial inulin concentration of 20 g/L  135
Table 5.27.3  Biomass concentration time history with initial inulin concentration of 30 g/L  135
Table 5.27.4  Biomass concentration time history with initial inulin concentration of 40 g/L  136
Table 5.28  Experimental values of specific cell growth rate at different initial concentration of inulin and glucose  138
Table 5.29  Simulated values of specific cell growth rate at different initial concentration of inulin and glucose  140
Table 5.30  Specific cell growth rate of *Pediococcus acidilactici* in presence of inulin isolated from wheat with concentration of glucose as parameter  146
<table>
<thead>
<tr>
<th>Table 5.31</th>
<th>Specific cell growth rate of <em>Pediococcus acidilactici</em> in presence of inulin isolated from oat with concentration of glucose as parameter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Table 5.32</td>
<td>Specific cell growth rate of <em>Pediococcus acidilactici</em> in presence of inulin isolated from dalia with concentration of glucose as parameter</td>
</tr>
<tr>
<td>Table 5.33</td>
<td>Biomass concentration time history with garlic paste at glucose concentration 20 g/L</td>
</tr>
<tr>
<td>Table 5.34</td>
<td>Specific cell growth rate of <em>Pediococcus acidilactici</em> as function of initial garlic paste concentration</td>
</tr>
<tr>
<td>Table 5.35</td>
<td>Experimental values of maximum specific cell growth rate and substrate saturation constant up to garlic paste concentration 120 g/L for glucose concentration 20 g/L.</td>
</tr>
<tr>
<td>Table 5.36</td>
<td>Experimental values of kinetic parameters present in equation</td>
</tr>
<tr>
<td>Table 5.37</td>
<td>Concentration of biomass grown under basil leaf extract medium at different time</td>
</tr>
<tr>
<td>Table 5.38</td>
<td>Specific cell growth rate of <em>Pediococcus acidilactici</em> at different concentration of basil leaf extract</td>
</tr>
<tr>
<td>Table 5.39</td>
<td>Numerical values of $\mu_m$ and $K_s$</td>
</tr>
<tr>
<td>Table 5.40</td>
<td>Time concentration history of <em>Pediococcus acidilactici</em> in presence of betel leaf extract solution</td>
</tr>
<tr>
<td>Table 5.41</td>
<td>Specific cell growth rate of <em>Pediococcus acidilactici</em> at different concentration of betel leaf extract</td>
</tr>
<tr>
<td>Table 5.42</td>
<td>Numerical values of $\mu_m$ and $K_s$</td>
</tr>
<tr>
<td>Table 5.43</td>
<td>Prebiotic score of different prebiotics against selected pathogens</td>
</tr>
</tbody>
</table>
Table 5.44  Zone of inhibition exhibited by probiotic after exposed to prebiotic at different time interval  163
Table 5.45  Weight percentage of different elements constituents in biosynthesised basil-silver nanoparticles  167
Table 5.46  Biomass concentration time history with different concentrations of basil-AgNPs  169
Table 5.47  Specific cell growth rate with different concentration of basil-AgNPs  171
Table 5.48  Numerical values of $\mu_m$ and $K_s$  171
Table 5.49  Weight percentage of different elements constituents in biosynthesised basil-silver nanoparticles  174
Table 5.50  Chemical groups present in betel-AgNPs  175
Table 5.51  Biomass concentration time history with different concentrations of betel-AgNPs  176
Table 5.52  Specific cell growth rate with different concentration of betel-AgNPs  177
Table 5.53  Numerical values of $\mu_m$ and $K_s$
### Appendix

| Table 8.A.1 | Biomass concentration time history with 5 g/L wheat-inulin | 186 |
| Table 8.A.2 | Biomass concentration time history with 10 g/L wheat-inulin | 186 |
| Table 8.A.3 | Biomass concentration time history with 15 g/L wheat-inulin | 187 |
| Table 8.A.4 | Biomass concentration time history with 20 g/L wheat-inulin | 187 |
| Table 8.A.5 | Biomass concentration time history with 5 g/L oat-inulin | 188 |
| Table 8.A.6 | Biomass concentration time history with 10 g/L oat-inulin | 188 |
| Table 8.A.7 | Biomass concentration time history with 15 g/L oat-inulin | 189 |
| Table 8.A.8 | Biomass concentration time history with 20 g/L oat-inulin | 189 |
| Table 8.A.9 | Biomass concentration time history with 5 g/L dalia-inulin | 190 |
| Table 8.A.10 | Biomass concentration time history with 10 g/L dalia-inulin | 190 |
| Table 8.A.11 | Biomass concentration time history with 15 g/L dalia-inulin | 191 |
| Table 8.A.12 | Biomass concentration time history with 20 g/L dalia-inulin | 191 |