Translating Simulation Approaches for Immunology

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Publications

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Abstract

This thesis presents a novel set of guidelines to convert between simulation modelling approaches, namely, Ordinary Differential Equations (ODEs), System Dynamics (SD) and Agent-based Modelling and Simulation (ABMS). In our literature review we identify a gap in establishing translation techniques between these approaches. We therefore focus our research in developing these techniques and assessing the impact of these conversions in the simulation outcomes. In particular, our interest lies in investigating our techniques applied to simulation problems for the immune system, as we wish to aid immunologists with the choice of the most appropriate approach for a certain problem.

The aims of this thesis are therefore defined as: (1) with no explicit guidelines available from the literature, we want to develop, test and validate our own set of guidelines for converting between approaches: from ODE models to SD, from SD to ABMS and from ABMS to SD; and (2) we seek to discuss the merits of SD and ABMS for immunology to assist researchers with the choice between both approaches.

The assessment of the effectiveness of the conversion guidelines is achieved by using a case study approach involving six cases of established mathematical models describing immunological phenomena. These case studies are chosen by considering aspects such as the behaviour of the entities of the model (whether they are static or interact with other entities and whether they have spatial representation or not), the type of hypothesis to be tested, the empirical embeddedness of real data, population sizes, number of elements involved and the modelling effort.

In order to conduct our conversion for the case studies, we first convert their original ODE model into an SD model, and then perform the translation from SD to ABMS. For the last three case studies, we also test the conversion guidelines from ABMS to SD. Evidence from the experiments reveal that for all cases it was possible to obtain equivalent approaches by using the conversion guidelines developed. However, outcome differences occur given the intrinsic characteristics of each simulation modelling paradigm. By observing these differences we could conclude that (1) SD is incapable of reflecting exactly the same variability as that obtained from the agent-based simulation, as it is a deterministic approach; (2) SD variables change continuously in time and therefore population numbers over time might be different from those obtained by the agent-based simulation; (3) as the number of different agents and behaviours increase, the corresponding SD becomes very intricate and difficult to develop and understand; (4) there are cases where it is preferable not to convert from ABMS to SD, as the agent-based model is easier to conceptualise and implement; (5) For other circumstances, ABMS outcomes are the same as those produced by the ODEs and SD, with the disadvantage to be more resource consuming in terms of computational memory and processing capacity; and (6) For some cases SD is less informative than ABMS, as it does not produce multiple scenarios or variations over the course of more than one run within the same parameters.

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Contents

1 Introduction **1**

1.1 Research Aim, Objectives and Contributions 3

1.1.1 Thesis Contributions 6

1.2 Simulation Experiments and Research Findings 6

1.3 Thesis Outline 8

2 Literature Review **9**

2.1 Introduction 9

2.2 The Theory of Simulation Modelling 10

2.2.1 System Modelling 10

2.2.2 Simulation 13

2.3 System Modelling and Simulation Methods 15

2.3.1 System Dynamics 15

2.3.2 Agent-based Modelling and Simulation 19

Cellular Automata (CA) 20

Agent 21

Agent-based Modelling Technique 23

2.4 Comparing Simulation Approaches 23

Simulation Modelling Abstraction Levels 25
2.5 Related Work 26
2.5.1 Contrasting SD and ABMS 26
2.5.2 Converting between Approaches 29
2.6 The Immune System 34
2.6.1 Humoral Immune Response 35
2.6.2 Cellular Immune Responses 37
Natural Killer Cells 37
B Cells 38
T Cells 38
Phagocytes 40
2.6.3 Immunosenescence 41
2.7 The Need for Simulation of the Immune System 42
2.8 Summary 45

3 Methodology **48**
3.1 Introduction 48
3.2 Research Problem 49
3.3 Research Aim, Objectives and Deliverables 50
3.4 Development of a Research Strategy 51
3.4.1 Principles of Research Design 51
3.4.2 Research Strategy 52
3.4.3 Developing our Conceptual Models 59
3.4.4 Experimentation and Validation 60

3.5 Conversion between Approaches 61
3.5.1 From Ordinary Differential Equations to System Dynamics . . . 61
Example 63
3.5.2 From System Dynamics to Agent-based Modelling and Simulation 65
Example 66
3.5.3 From Ordinary Differential Equations to Agent-based Modelling and Simulation 69
Example 70
3.5.4 From Agent-based Modelling and Simulation to System Dynamics 71
Example 73
3.6 Summary 76

4 Static Non-spatial models **77**

4.1 Introduction 77
4.2 Case 1: Naive T Cells Output 78
4.2.1 Lack of naive T cells 79
4.2.2 Naive T cell output 79
4.2.3 The Mathematical Model 80
4.2.4 The System Dynamics Model 83
From Ordinary Differential Equations to System Dynamics . . . 83
Model Stocks 84
Model Flows 84
Model Information 86
Model Parameters 87
Flows Calculations 88

The Final System Dynamics Model 88
4.2.5 From System Dynamics to Agent-based Modelling and Simulation 90
Model Agent 90
Agent Behaviours 91
Agent Implementation 92
Simulation 93
4.2.6 Experiments 93
4.2.7 Results 97
4.2.8 Summary of Case 1 102
4.3 Case 2: Tumour Growth 104
4.3.1 Mathematical Models 104
4.3.2 The System Dynamics Model 106
From Equations to System Dynamics 106
Model Stocks 106
Model Flows 106
Model Information 107
Model Parameters 107
Flows Calculations 107
The Final System Dynamics Model 107
4.3.3 From System Dynamics to Agent-based Modelling and Simulation 108
Model Agent 108
Agent Behaviours 109
Agent Implementation 109
Simulation 110

4.3.4 Experiments . . . 110
4.3.5 Results . . . 111
4.3.6 Summary of Case 2 . . . 115
4.4 Case 3: Viral Spread of Human Immunodeficiency Virus . . . 118
4.4.1 Mathematical Model . . . 118
4.4.2 The System Dynamics Model . . . 120
From Ordinary Differential Equations to System Dynamics . . . 120
Model Stocks . . . 120
Model Flows . . . 120
Model Information . . . 122
Model Parameters . . . 123
Flows Calculations . . . 123
The Final System Dynamics Model . . . 124
4.4.3 From System Dynamics to Agent-based Modelling and Simulation 124
Model Agents . . . 124
Agents' Behaviours . . . 125
Agents' Implementation . . . 126
Simulation . . . 128
4.4.4 Experiments . . . 128
4.4.5 Results . . . 128
4.4.6 Summary of Case 3 . . . 136
4.5 Summary . . . 136

5 Dynamic Spatial and Non-Spatial Models 139

5.1 Introduction 139

5.2 Case 4: Interactions between Tumour Cells and Generic Effector Cells . 140

5.2.1 The Mathematical Model 141

5.2.2 The System Dynamics Model 143

From Ordinary Differential Equations to System Dynamics . . . 143

Model Stocks 143

Model Flows 143

Model Information 144

Model Parameters 145

Flows Calculations 145

The Final System Dynamics Model 145

5.2.3 From System Dynamics to Agent-based Modelling and Simulation 145

Model Agents 145

Agents' Behaviours 146

Agent Implementation 147

Simulation 148

5.2.4 Experiments 148

5.2.5 Results 149

5.2.6 Summary of Case 4 – Converting from System Dynamics to Agent-Based Modelling and Simulation 156

5.2.7 From Agent-based Modelling and Simulation to System Dynamics 158

5.2.8 From Agent-based Modelling and Simulation to System Dynamics Considering Agents' Movement 162

The Agent-based Model 163

System Dynamics Model for the Dynamic cells of the Agent-based Model . . . 163
5.2.9 Experiments . . . 164
5.2.10 Results for the Static Model . . . 165
5.2.11 Results for the Dynamic Model . . . 165
5.2.12 Summary of Case 4 – Converting from Agent-Based Modelling and Simulation to System Dynamics . . . 165
5.3 Case 5: Interactions Between Tumour Cells, Effector Cells and Cytokines IL-2 . . . 170
5.3.1 The Mathematical Model . . . 170
5.3.2 The System Dynamics Model . . . 171
From Ordinary Differential Equations to System Dynamics . . . 171
Model Stocks . . . 171
Model Flows and Information . . . 171
Model Parameters . . . 172
Flows Calculations . . . 172
The Final System Dynamics Model . . . 172
5.3.3 From System Dynamics to Agent-based Modelling and Simulation 174
Model Agents . . . 174
Agents' Behaviours . . . 174
Agent Implementation . . . 174
Simulation . . . 175
5.3.4 Experiment . . . 176
5.3.5 Results . . . 176

5.3.6 From Agent-based Modelling and Simulation to System Dynamics 180
The Agent-based Model . . . 180
System Dynamics Model . . . 181
Experiment . . . 182
Results . . . 183
Summary of Case 5 . . . 187
5.4 Case 6: Interactions Between Tumour Cells, Effector Cells, IL-2 and TGF-β . . . 188
5.4.1 The Mathematical Model . . . 188
5.4.2 The System Dynamics Model . . . 190
Model Stocks, Flows and Information . . . 190
Model Parameters . . . 191
Flows Calculations . . . 191
The Final System Dynamics Model . . . 192
5.4.3 From System Dynamics to Agent-based Modelling and Simulation 192
Model Agents . . . 192
Agents Behaviours . . . 192
Agent Implementation . . . 193
5.4.4 Experiment . . . 193
5.4.5 Results . . . 193
5.4.6 Summary of Case 6 – Converting from System Dynamics to Agent-based Modelling and Simulation . . . 205
5.4.7 From Agent-based Modelling and Simulation to System Dynamics 206
The Agent-based Model . . . 206

System Dynamics Model 207
Experiment 207
Results 207
Summary of Case 6 – Converting from Agent-based Modelling and Simulation to System Dynamics 213
5.5 Summary 213

6 Conclusions **217**

6.1 Introduction 217
6.2 Evaluation of Aims 219
Experimentation 220
6.3 Research Findings 221
6.4 Limitations of the Study 224
6.5 Future Work 225

List of Tables

1.1 Case studies 5

2.1 Main differences between SD and ABMS (obtained from [76]) 25

3.1 Flow calculations for the predator-prey example 64

3.2 SD flows converted into ABMS 68

4.1 Rate values for the mathematical model (obtained from [59]) 83

4.2 Parameters from the mathematical model and their correspondents from the SD model 88

4.3 Flow calculations for the naive T cell output model 89

4.4 Agents' parameters and behaviours for the naive T cell output model . . 91

4.5 Simulation parameters for different scenarios. The parameter c is only used in the first scenario, where there is no proliferation. In the other scenarios, proliferation is defined by the equation $\left(1 + \frac{300}{N_p}\right) * \frac{4.4}{h}$ [59]. . 94

4.6 The data set used for validation obtained in [59] and [15] 95

4.7 The data set collected in Lorenzi *et al.* [50] 96

4.8 Wilcoxon test with 5% significance level comparing the results from SD and ABMS for the data sets used for validation on the number of naive T cells from thymus 101

4.9 Flow values calculations 108

4.10 Agents' parameters and behaviours for the tumour growth model 109

4.11 Transition rate calculations from the SD flow equation for the tumour growth model 110

4.12 Wilcoxon test with 5% significance level comparing the results from SD and ABMS for the Tumour Growth model. The hypothesis of similarity for most cases was rejected ($p \simeq 0$) 111

4.13 Parameters from the mathematical model, their correspondents and values for the case 3 SD model 123

4.14 Flow calculations for the viral spread model 124

4.15 Agents' parameters and behaviours for the viral spread model 126

4.16 Transition rates calculations from SD flows equations for the viral spread model 127

5.1 Flow calculations for case 4 146

5.2 Agents' parameters and behaviours for case 4 146

5.3 Transition rates calculations from SD flows equations for case 4 148

5.4 Simulation parameters for different scenarios. For the other parameters, the values are the same in all experiments, i.e. $a = 1.636$, $g = 20.19$, $m = 0.00311$, $n = 1$ and $p = 1.131$, as defined in [45]. The four scenarios investigate variations in the aggressiveness of the tumour (parameters a and b), apoptosis of effector cells (parameter d) and amount of cells injected as treatment (s). p and g define the proliferation of effector cells and n determines the rate in which effector cells kill tumour cells. 149

5.5 Wilcoxon test comparing case 4 simulation results 150

5.6 Transition rates from ABMS model for case 4 161

5.7 Flow calculations for case 4 from ABMS to SD 161

5.8 Simulation parameters for different scenarios in the dynamic models. For the other parameters, the values are the same in all experiments, i.e. $a = 1.636$, $g = 20.19$, $m = 0.00311$, $n = 1$ and $p = 1.131$. 164

5.9 Flow calculations for case 5 173

5.10 Agents' parameters and behaviours for case 5 174

5.11 Transition rates calculations from SD flows equations for case 5 175

5.12 Parameter values for case 5 176

5.13 Wilcoxon test with 5% significance level comparing the results from SD and ABMS for case 5 180

5.14 Flow calculations for case 6 191

5.15 Agents' parameters and behaviours for case 6 193

5.16 Parameter values for case 6 195

5.17 Transition rates calculations from SD flows equations for case 6 196

6.1 Summary of findings with regards to the conversion from SD to ABMS . 222

6.2 Summary of findings with regards to the conversion from ABMS to SD . 223

List of Figures

2.1 Filling a glass of water problem [41] 16

2.2 Filling a glass of water problem: Causal loop diagram [41] 17

2.3 Filling a glass of water problem: Stock and flow diagram 18

2.4 Filling a glass of water problem: SDS 19

2.5 Examples of classical neighbourhoods in a two-dimensional grid CA . . 20

2.6 An agent (obtained from [53]) 22

2.7 Dendritic cell state chart. The round squares represent the states and the arrows represent the transitions between the states. Arrows inside states indicate the agent actions (or behaviours). 24

2.8 Correspondence of SD and ABMS: Constant and Proportional rates (obtained from [8]). 31

2.9 Correspondence of SD and ABMS: Multiple Stocks and Compositionality (obtained from [8]). 32

2.10 Primary and secondary immune responses (obtained from [87]). 36

2.11 The antibody (obtained from [37]). 39

2.12 Phagocytes (obtained from [37]). 40

3.1 Process of simulation study: steps and their iterations. 59

3.2 Example of information definition 62

3.3 The stock x and its flows and information 64

3.4 The stock y and its flows and information. 64

3.5 The final SD model for the predator-prey example 65

3.6 The SD model for bass diffusion 67

3.7 The bass diffusion agent 69

3.8 The bass diffusion agent 71

3.9 The SIR agent 73

3.10 The SD model stocks and flows for the SIR model 74

3.11 The SD model stocks and flows and information for the SIR model . . . 74

3.12 The final SD for the SIR model 76

4.1 Dynamics of Naive T cells 80

4.2 The data set used as a look-up table for the active cells. The data set contains the number of activated $CD4$ cells per mm^3 for early years taken from Comans-Bitter *et al.* [14]. 84

4.3 The naive T cell stock variable and its flows: thymic output, proliferation and death 85

4.4 The naive T cells from proliferation stock variable and its flows: death, activation, proliferation and memory reverted to naive 85

4.5 The memory T cells stock variable and its flows: reversion to a naive phenotype, reversion from active and death 86

4.6 The naive T cell stock variable with its flows and information 87

4.7 The naive T cells from proliferation stock variable with its flows and information 87

4.8 The memory T cells stock variable with its flows and information 87

4.9 The system dynamics model's functions and parameters for case 1. The table function RealActives returns the values for active CD4+ cells at a certain time. 89

4.10 T cell agent states 91

4.11 The T cell agent 92

4.12 New agent (T cell) state decision flow chart 93

4.13 Data sets used for validation of the naive T cell output simulation models 96

4.14 Results for naive T cells from thymus 98

4.15 Results for naive T cells from peripheral proliferation 99

4.16 Results for total T cells 100

4.17 The tumour cells stock variable and its flows: proliferation and death . 106

4.18 The tumour cells stock variable and its flow: $ProliferationMinusDeath$ 106

4.19 The tumour cells stock variable with its flow and information 107

4.20 System dynamics model for the one-equation mathematical model . . . 108

4.21 Tumour Cell agent 109

4.22 Results for the tumour growth model varying the c parameter: Logistic model 112

4.23 Results for the tumour growth model varying the c parameter: Von Bertalanffy model 113

4.24 Results for the tumour growth model varying the c parameter: Gompertz model 114

4.25 Results for the second experiment of one-equation logistics model using SD and ABMS 116

4.26 Results for the second experiment of tumour growth Von Bertalanffy (a) and Gompertz (b) models using SD 117

4.27 The healthy CD4 cells stock and its flows: source (*Source*), cellular growth *Growth*), death (*HealthyCD4Death*) and infection (*Infection*) 121

4.28 The infected CD4 cells stock and its flows: source (*SourceInfected*) and death (*deathInfected*) 121

4.29 The free HIV stock and its flows: source (*SourceFreeHIV*), infection of CD+ cells (*Infection*) and viral death (*deathHIV*) 121

4.30 The *HealthyCD4* stock with its flows and information 122

4.31 The *InfectedCD4* stock with its flows and information 122

4.32 The *FreeHIVConcentration* stock with its flows and information . . . 123

4.33 The system dynamics model's functions and parameters for case 3 . . . 125

4.34 The CD4+ T cell agent state chart 126

4.35 The free HIV agent state chart 127

4.36 The free HIV agent state chart with constraint 129

4.37 ODE, SD and ABMS results for the healthy CD4+ T cells 130

4.38 ODE, SD and ABMS results for the actively infected CD4+ T cells . . . 131

4.39 ODE, SD and ABMS results for the HIVs 132

4.40 SD and ABMS closest results for the healthy CD4+ T cells 133

4.41 SD and ABMS closest results for the actively infected CD4+ T cells . . 134

4.42 SD and ABMS closest results for the HIVs 135

5.1 The tumour cells stock variable and its flows: *proliferationMinusdeath* and tumour cells *killedbyEffectorCells* 143

5.2 The effector cells stock variable and its flows: proliferation, death, apoptosis and treatment 144

5.3 The tumour cells stock variable with its flows and information 144

5.4 The effector cells stock variable with its flows and information 145

5.5 SD model for case 4 147

5.6 ABMS state charts for case 4 147

5.7 Results for scenario 1 152

5.8 Results for scenario 2 153

5.9 Results for scenario 3 154

5.10 Results for scenario 4 155

5.11 Results for case 4. In the first column the ODE results for the four scenarios are shown. The second column presents the SD results and the third column presents the ABMS results. The fourth and fifth columns show the fixes attempted to achieve similar outcomes. Fix 1 adds the constraint $tumourcells > 0$. Fix 2 adds the constraint $tumourcells > 0$ and $effectorcells > 0$. Each graph illustrates the results for tumour cells (continuous line) and effector cells (dotted lines). The scale for the results for tumour cells is on the left y axis of each graph while the scale for effector cells is on the right y axis. 157

5.12 ABMS state charts for conversion to SD 159

5.13 SD stock and flow diagram 160

5.14 SD model 162

5.15 ABMS state charts for case 4 considering agent's movement 163

5.16 SD model corresponding to the dynamic agent-based model 164

5.17 Results for scenario 1 considering cellular movement 166

5.18 Results for scenario 2 considering cellular movement 167

5.19 Results for scenario 3 considering cellular movement 168

5.20 The effector cell stock variable with its flows and information 171

5.21 The IL-2 stock variable with its flows and information 172
5.22 SD model for case 5 173
5.23 ABS state charts for the agents of case 5 175
5.24 SD and ABMS results for effector cells 177
5.25 SD and ABMS results for tumour cells 178
5.26 SD and ABMS results for IL2 179
5.27 SD model corresponding to the dynamic agent-based model for case 5 . 183
5.28 Results for effector cells considering cellular movement 184
5.29 Results for tumour cells considering cellular movement 185
5.30 Results for IL-2 considering cellular movement 186
5.31 Stocks, flows and information for case 6 190
5.32 SD model for case 6 192
5.33 ABS state charts for the agents of case 6 194
5.34 ODE, SD and ten runs of ABMS results for effector cells 197
5.35 ODE, SD and ten runs of ABMS results for tumour cells 198
5.36 ODE, SD and ten runs of ABMS results for IL-2 199
5.37 ODE, SD and ten runs of ABMS results for TGF-β 200
5.38 SD and ABMS results for effector cells 201
5.39 SD and ABMS results for tumour cells 202
5.40 SD and ABMS results for IL-2 203
5.41 SD and ABMS results for TGF-β 204
5.42 SD model corresponding to the dynamic agent-based model for case 6 . 208
5.43 Results for effector cells considering cellular movement for case 6 209
5.44 Results for tumour cells considering cellular movement for case 6 210

5.45 Results for IL-2 considering cellular movement for case 6 211

5.46 Results for TGF-β considering cellular movement for case 6. As ABMS is discrete and the values for the TGF-β in the model are smaller than zero, they do not appear in the outcome. 212

List of Abbreviations

ABMS - Agent-based modelling and simulation

CA - Cellular Automata

IS - Immune system

ODE - Ordinary differential equations

SD - System dynamics

SDS - System dynamics simulation

Chapter 1

Introduction

Although there are examples of successful applications of modelling and systems simulation approaches to immunology, these are not commonly used to assist immunological research. Instead, ordinary differential equation (ODE) models are widely employed to support advances in theoretical immunology. The development of an ODE model, however, requires that the immune model developers have an in depth understanding of mathematics, which is not the immunologists main area of expertise. Furthermore, these models have limitations for solving complex problems which involve individual localisation, memory and emerging properties. Systems simulation is therefore a set of methodologies that has emerged to complement mathematical models and overcome some of their limitations. Possible systems modelling and simulation approaches for immunology include system dynamics (SD) and agent-based modelling and simulation (ABMS).

SD encompasses the ODEs' mathematical formulations with the advantage of modelling the system as a stock and flow diagram, which seemed more intelligible by the immunologists that we worked with during the development of our models. Furthermore, there is no need for an indepth understanding of mathematics to formulate a model. The differential equations are implicit in the system's structure and the relationships between the elements modelled can be established with experimental data.

ABMS modelling approaches, compared to ODEs and SD models, are acknowledged as more suitable for simulations in which there is the need to represent individual cells, their interactions and emergent behaviour.

There are several reasons why systems modelling and simulation are not as common as ODEs for immunology: (1) System simulation tools are not known in the immunology research field; (2) although system simulation is acknowledged as being a useful tool by immunologists, there is no knowledge of how to use it; and (3) there is not enough trust in the results produced by simulation. One way of overcoming the unpopularity of systems simulation in immunology would be re-conceptualizing or converting existing established ODE models of immune processes into a systems simulation perspective.

In this thesis, therefore, **we seek to define guidelines for the conversion between ODEs, SD and ABMS for immune system problems that can benefit from either approach**. To our knowledge, there is little research concerned with the translation between these approaches for immunology. We decided to apply our conversion guidelines to case studies in immunology for two main reasons. First, for the simulation of immune problems, apart from ODEs which have been widely explored, both SD and ABMS approaches have the potential to be very useful and can overcome some the ODE modelling limitations. Moreover, we had the opportunity to work closely with immunologists from Nottingham City Hospital and observe their acceptance of our SD and ABMS models contrasted to the ODE models.

Another possible advantage of the conversion guidelines would be in circumstances where a model developer is well acquainted with one simulation approach and would like to use his current models to learn another technique. In cases where there is, for example, a established ODE model, conversion techniques to translate the current model to ABMS would be a good starting point to learn this simulation approach and possibly expand the model.

Furthermore, immunology is a field that constantly gathers new information. Simulations therefore have to be updated frequently to suit new findings. For some cases, in

order to meet new demands, the replacement of the current simulation approach for new developments needs to be considered. There are two options, namely, to start the new simulation from scratch or to adapt previous implementations for the new approach. However, with regard to the second option, there is also little knowledge in literature on the implications of these conversions.

In addition, in a range of immune problems that are solvable by the three approaches, apart from their well-known intrinsic characteristics, it is necessary to identify aspects such as (1) the available data, (2) the time given to build the model, (3) the research questions to be addressed, (4) the possible knowledge to be acquired regarding the real-world system, (5) the modelling effort and (6) the computational resources available. It might be the case where the available approach that better matches the resources constraint is not well known by the simulation developer. However, establishing a parallel between what could be done in the preferable approach and the available technique should facilitate the model building. Furthermore, with regard to multi-paradigm simulation, in order to make model simplifications or decrease the demand for computational resources, there is the need to establish whether one simulation approach can be replaced by another without any detriment to the model validity. **We also want to investigate, therefore, what are the outcome impacts of converting approaches and those circumstances where such effort should be considered**.

1.1 Research Aim, Objectives and Contributions

Taking into account the research gap presented int the previous section, we outline our research aims as follows:

- With no explicit guidelines available from the literature, we aim to develop, test and validate our own set of guidelines for converting between ODE, SD and ABMS approaches. We believe these practices will assist with the improvement and expansion of existing immune models to suit new demands.

- We seek to asses the impacts of these conversions and determine those circumstances where one approach could be replaced by another. We also want to determine, after performing the translation, when one approach would be preferable than the other.

In order to achieve our aims, the main objectives of our work are:

1. To define and test guidelines to convert between (1) ODE models to SD models, (2) ODE models to ABMS models (3) SD models to ABMS models, (4) ABMS models to SD models and to assess the impact of these conversions.

2. To compare ODE, SD and ABMS outcomes, considering aspects such as the behaviour of the entities of the model (whether or not they are static or interact with other entities, and whether they have spatial representation or not), the type of hypothesis to be tested and the modelling effort.

3. To define guidance to choose between SD and ABMS depending on the characteristics of the problem to be addressed.

In order to achieve these aims, several case studies of established mathematical models that describe immune mechanisms are investigated. These case studies are chosen by considering aspects such as the behaviour of the entities of the model (whether they are static or interact with other entities and whether or not they have spatial representation), the type of hypothesis to be tested, the empirical embeddedness of real data and the modelling effort. Furthermore, we consider characteristics such as population size, model complexity, observation of the ODEs outcome results (when there is no data available) and the number of different populations modelled. The mathematical models chosen vary largely within these aspects and therefore we can perform a more robust analysis on the effectiveness of our guidelines. In addition, we select models relating to topics that currently are being investigated by the immunologists from Nottingham City Hospital, including those involved with acquired immunodeficiency syndrome (AIDS),

cancer and immune system ageing. We define SD and ABMS models based on the scenarios described by these mathematical models, and convert and compare the simulation results.

Table 1.1: Case studies

Case Study	Entities	Movement	Data available	Pop. sizes	Num. of pop.	Complexity
1) Naive T cell output	static	no	yes	10^3	1	medium
2) Tumour growth	static	no	no	10 to 10^{70}	1	low
3) HIV spread	interact	no	no	10^4	3	medium
4) Tumour/Effector	interact	yes	no	6 to 600	2	low
5) Tumour/Efector/ IL-2	interact	yes	no	10^4	3	medium
6) Tumour/Effector/ IL-2/TGF-β	interact	yes	no	10^4	4	high

The first case study – which is a complex model in terms of population sizes and elements considered – is based on an ODE model involving interactions that influence naive T cell populations with age (Section 4.2). For the second case study – which is the simplest one with only one population – we investigate mathematical models of general tumour growth (Section 4.3). Our third case study – which also complex in terms of different populations – comprises an ODE model of cell-free viral spread of the human immunodeficiency virus (HIV) in the bloodstream (Section 4.4). The fourth case study is based on an ODE model involving interactions between tumour cells and generic effector cells (Section 5.2). The fifth case study adds to the previous model the influence of IL-2 cytokine molecules in the immune responses of effector cells towards tumour cells (Section 5.3). The final case study comprises an ODE model of interactions between effector cells, tumour cells, and IL-2 and TGF-β molecules (Section 5.4). For the last three case studies we considered spatial movement. We believe these new requirements will provide means to further test our conversion guidelines. We also hypothesise that spatial interactions are more suitable for the ABMS models and their conversion to an equivalent SD might result in a rather complex stock and flow diagram.

1.1.1 Thesis Contributions

As contributions to knowledge, we define and test our conversion guidelines, compare simulation results of case studies, and identify their characteristics that allow for the production of similar results for both modelling efforts. In addition, when the simulation results differ, we investigate why these differences occur and if they are systematic. Moreover, we discuss the merits of each simulation approach considering the modelling effort, simulation performance -in terms of accuracy and computational resources used- and experimental data available.

1.2 Simulation Experiments and Research Findings

In our first set of experiments, we convert between approaches and compare a SD model and an ABMS model for an immune system ageing problem. The problem involves a non-spatial model with static agents. By static, we mean that there is no movement or interaction between the agents. We obtain the same simulation results for both approaches. If we consider aspects such as modelling effort, necessary level of abstraction and the experimental data available, however, our results indicate that for these types of individual entities, SD modelling is preferable. Moreover, when contrasting the simulation results for both modelling efforts in our first case study, the SD simulation is less complex and takes up fewer computational resources, producing the same results as those obtained by the ABMS. In addition, SD simulation is more robust when the number of individual entities increases considerably. There were cases in which there were insufficient computational resources to run the ABMS in the machine our experiments were run.

As a second set of experiments, we investigate mathematical models of tumour growth and obtain close, but not exactly the same results, in both approaches to simulation. As agents are integer values, the growth curve for the ABMS is more accentuated than that from SD. Moreover, similar to the previous experiments, SD is more suitable in

terms of scalability of the number of tumour cells.

Another case study investigates the spread of the human immunodeficiency virus (HIV) and its interactions with immune cells. Although it is possible to convert between approaches, the stochastic character of the ABMS produces a range of results with the same trend as those from the SD, but with growth and decrease of populations starting at different points in time for each run.

The last three case studies are concerned with general interactions between immune effector populations and tumour cells. Some scenario results indicate that the size of the populations also influence the similarity of results, as a small number of agents can produce significant variability in the outcomes. Furthermore, another result suggests that extra efforts, such as parameter calibration, are necessary to obtain equivalent results for both approaches after applying the conversion techniques. In addition, there is a final case study in which the outcomes for the ABMS mostly follows the same pattern as that produced by the SD; however there are some alternative outcomes other than that produced by the deterministic models. This indicates that for this case study, the ABMS results show other possible population dynamics that should be validated by immunologists.

Based on our experiments, it was not possible to define a general framework that would definitively determine the most suitable approach depending on the problem investigated. We could only establish some rules that indicate when one approach is preferable than the other by observing characteristics such as population sizes, whether the simulation regards continuous or discrete values, the entities spatial movement and the representation of different populations in the problem. However these rules are specific for our set of case studies and further investigation would be necessary to determine whether they can be generalized.

1.3 Thesis Outline

This thesis is structured in the following manner. Chapter 2 presents the relevant background information and is divided into seven sections: the first introduces the main concepts of modelling and simulation; secondly, the main system simulation approaches used in this work are described; the third and fourth sections review the literature regarding the use and comparison of ABMS with SD; the fifth section reviews the main concepts of immunology necessary to understand the case studies; section six investigates the opportunities for applying simulation to immunology, followed by the summary of the chapter, which outlines the research gaps found in literature. Chapter 3 presents the methodology used to conduct our case studies experimentation and introduces the conversion guidelines that we have defined. Chapter 4 comprises three case studies regarding non-spatial entities within the system, with the objective of testing our guidelines. Chapter 5 reports on further tests on our guidelines by investigating their effectiveness when applied to spatial models. The thesis concludes with a discussion of findings, an assessment of how far aims have been satisfied and future directions for this research (Chapter 6).

Chapter 2

Literature Review

2.1 Introduction

The research described in this thesis is concerned with the translation between the modelling and simulation paradigms ODEs, SD and ABMS. It investigates the means to convert from one approach to another and assess the consequences of the conversions in terms of model performance, complexity and output accuracy. The research is conducted with particular attention to case studies in immunology, as this is an area in which the potential contributions of simulation have not been largely explored.

This chapter starts by introducing the main concepts of the theory of modelling and simulation, and presents established definitions for systems and their dominant features. Furthermore, definitions of a model as well as the major classes and dimensions of system models are provided. Subsequently, the principal concepts of model simulation are introduced in Section 2.2.

Section 2.3 describes each simulation approach used in this thesis. It focuses on their methodology, modelling technique, architecture and applicability. We review SD and ABMS. Furthermore, each methodology is compared against each other to outline the main differences between them with respect to their building blocks, level of abstraction, time handling, outcomes, perspectives, etc.

As the focus of this research is to establish a framework to convert between SD and ABMS, studies concerned with the comparison and conversion of these methodologies are reviewed in Section 2.5. Furthermore, the limitations of the current efforts to convert between methods and their potential for improvement are discussed.

Section 2.6 reviews the key concepts of immunology, focusing on the main processes, cells and tissues of the immune system. These concepts will be further used in the remainder of this chapter and for our case studies in subsequent chapters. Section 2.7 investigates the opportunities of applying simulation in immunological research and justifies the choice of immunology case studies adopted in this thesis. Finally, Section 2.8 provides a summary of the chapter.

2.2 The Theory of Simulation Modelling

In this section we review the main concepts concerned with the theory of systems modelling and simulation. First, we present a definition of a system and its principal characteristics. Subsequently, we explain concepts related to system modelling and model simulation.

2.2.1 System Modelling

A system can be defined as a "*collection of parts organized for some purpose*" [16]. There are four main classes of systems [12]:

1. Natural systems: systems whose origins lie in the origins of the universe. For example, atoms, molecules and galactic systems.

2. Designed physical systems: physical systems designed by humans. For example, cars, mobile phone networks and computers.

3. Designed abstract systems: abstract systems designed by humans. For example, mathematical models and literature.

4. Human activity systems: systems of human activity that are consciously or unconsciously ordered. For example, family, schools, cities and criminal justice systems.

All these systems can be modelled and simulated [70]. In this thesis we consider only natural systems and designed abstract systems. Furthermore, in some cases, we use designed abstract systems to describe natural systems. Our final systems, therefore, lie at the interface between natural and designed abstract systems.

In order to study a system, experimentation can be done either over the actual system or a system model. The construction of a system model starts with understanding the real world. This allows for the characterisation of the system problems within the context of real-world observations. After the system is properly described, it is possible to conceptualize the model by defining its scope, objectives, inputs, outputs and simplifications [70].

There are different definitions of a system model. Fishman [29] defines model as "*a formal representation of theory or a formal account of empirical observation*". According to Banks [4], "*a model is a representation of an actual system. The model should be complex enough to answer the questions raised, but not too complex*". For Zeigler [99], a model is "*a set of instructions, rules, equations or constraint for generating input/output behaviour*". Kelton [39] characterizes a model as "*a set of approximations and assumptions, both structural and quantitative about the way the system does or will work*".

Although the authors offer different definitions for a model, they agree that modelling is "*creating an abstraction of the real world system using modelling tools with the objective of solving a problem*".

Once the real world problem is well understood, there is the need for the definition of the best modelling approach. There is a wide spectrum of modelling methodologies, which range from analytical modelling to simulation modelling.

Analytical models, also known as static models, consist of a set of mathematical equations that describe relationships among variables. The objective of using these equations is to approximate or predict the system behaviour [8]. These models rely on mathematical formulations to define the system's inputs and determine the outputs. Further, they focus on the determination of the system outcome given a certain input. Analytical models are not suitable to solve complex system problems as the solution might not exist or is very hard to find [8]. In addition, they do not give insights into the dynamics of the system as the analysis can only be done with a snapshot in time [79].

Simulation models emerged as a complement of analytical and numerical methods in order to model complex systems [51]. They are more appropriate for modelling dynamic and transient effects [63] and are well established [79]. Moreover, simulation models were reported as the second most important quantitative modelling technique, with statistics being the first [58].

Simulation models can be classified into four different dimensions [5, 39]. The first dimension is their representation concerned with time – they can either be static or dynamic [80]. Static simulation models represent a system at a particular point in time. Monte Carlo simulation models, for instance, are static [97]. Dynamic simulation models represent a system that evolves over time [2], such as simulation models for molecular interactions [77, 90].

The second dimension classifies simulation models into deterministic or stochastic models. Deterministic simulation does not contain any probabilistic components, such as the numerical simulation of a system of ordinary differential equations. Stochastic simulation, on the other hand, considers random components [5]. A simulation model of viral spread in a city, for example, is stochastic. It relies on the interactions between the individuals in the city, which are aleatory.

The third dimension establishes patterns in which system variables change over time. The variables can therefore change continuously or discretely. For example the age of an individual changes continuously in time, whereas the number of immune cells that

die with age is a discrete value [24].

The fourth dimension classifies the simulation models according to their demand for computational resources. Hence, simulation models can either be local (using one computer) or distributed (using more than one computer connected through a network) [2].

In the next section (Section 2.2.2) we present a review on the simulation basic concepts.

2.2.2 Simulation

A computational simulation of a dynamic system can be defined as an "*imitation (on a computer) of a system as it progresses through time*" [70].

The purpose of simulation, according to Pidd [64], is to understand, change, manage and control reality. Moreover, simulation can be used to obtain a better understanding of the system and/or to identify improvements to a system [70]. Another feature of simulation models is that they are focused on the main aspects of the real system. The models, therefore, are a simplified version that excludes unnecessary details of the original system [81].

A simulation predicts the performance of a system given a specific set of inputs. It is not the purpose of a simulation model to provide optimum or sub-optimum answers. According to Robinson [70], simulation is an experimental approach to modelling a "*what-if*" analysis tool. The model user determines the scenarios and the simulation predicts the outcomes. Simulation can, therefore, also be seen as a decision support tool.

There are three characteristics of system model (variability, interconnection and complexity) which allow for simulations capable of predicting a system performance, and of comparing different scenarios, designs and their impact on the outcomes produced [47].

Variations in the system can be predictable or unpredictable. For instance, if the death rate of an specific population of cells is known, as well as its initial value, it is possible to predict the variation of the number of these cells over time [59]. There are other

kinds of variations that cannot be predicted, for instance, if the same population of cells grows and decreases depending on the number of infections in the organism at a certain moment.

If a system is interconnected, their components do not work in isolation and affect each other. It is often difficult to predict the effects of interconnection, especially when there is variability. For instance, if the size of a population of immune cells influences the production of molecules that neutralizes toxins, when the immune cell population decreases, the molecule population will not grow and the toxins will therefore harm the organism [37, 59]; this often occurs with the ageing of the immune system [33, 55].

Another characteristic of many systems is complexity. There are two kinds of complexity to be considered: combinatorial and dynamic. Combinatorial complexity is related to the number of components in a system or the number of possible combinations of its components. Dynamic complexity, on the other hand, arises from the interactions of components in a system overtime and it is therefore not necessarily related to time [70].

Compared to real-world experimentation, simulation is generally more cost-effective and less time consuming. Furthermore, under a controlled simulation environment, changes and different scenarios are analysed without interfering in the real-world system operations [70]. Furthermore, as time can be accelerated in simulations, the impact of changes and the outcomes produced is many times faster than in the real world.

Another benefit of simulation is the possibility of testing systems that do not exist [81]. Such systems might not exist because they would be expensive or because direct experimentation is impossible. An abstract system, therefore, can be implemented and evaluated in a simulation environment.

One important issue during the development of a simulation model is the choice of the appropriate simulation method [6, 26, 49]. Current major system simulation modelling methods consist of system dynamics (SD) and agent-based modelling and simulation (ABMS) [8]. These methods will be explained in the next section.

2.3 System Modelling and Simulation Methods

In this section we introduce the systems simulation modelling techniques used: SD [31] and ABMS [53, 54]. In order to understand the differences and applicability of each approach, we review their main concepts, architecture, methodology and modelling technique. We also present a comparison between these methods. Furthermore, we discuss simulation abstraction levels, which indicate the level of detail that can be implemented by each approach.

2.3.1 System Dynamics

System Dynamics (SD) is a modelling methodology conceived by Jay Forrester in the mid 1950s [66]. It is an aspect of systems theory that was initially applied in order to understand complex aggregate behaviours in industry. It was, therefore, defined as "*the study of information feedback characteristic of industrial activity to show how organizational structure, amplification (in policies) and time delay (in decision and action) interact to influence the success of enterprise*" [31]. It is currently applied to any complex system characterized by interdependency, mutual interaction, information feedback and circular causality.

The basis of the SD methodology is the recognition that the structure of a system is just as important in determining its behaviour as the individual components themselves. It is therefore necessary to adopt a "systemic way of thinking" [41]. Systems thinking theories state that the shift from event orientation to focusing on the internal system structure increases the possibility of improving the system's performance. This means that the problem should be depicted as a set of patterns, interrelated processes and generic structures [68].

The main elements necessary to use SD methods are [32, 68]:

- Understanding the real-world dynamics over time

- Identifying the behaviour of the system entities at an aggregate level
- Regarding all the concepts from the real system as continuous values interconnected by loops of information feedback and circular causality (feedback loops)). Richardson and Pugh [68] define a feedback loop as "*a closed sequence of cause and effects, that is, a closed path of action and information*"
- Identifying the values that accumulate with time (stocks) in the system as well as their inflows and outflows
- Determining the causal/feedback structure of the system
- Defining the mathematical equations ruling the causal relationships between the stocks

In order to illustrate how these main elements are used, let us consider the problem of filling a glass of water from a tap [41]. We have a goal to seek, i.e. the desired level of water in the glass. To fill the glass with water, it is necessary to adjust the faucet position from the tap to control the water flow. This flow can be increased or decreased to achieve the desired level and its up to the user of the tap to decide this. The structure of the system can therefore be defined, as shown in Figure 2.1:

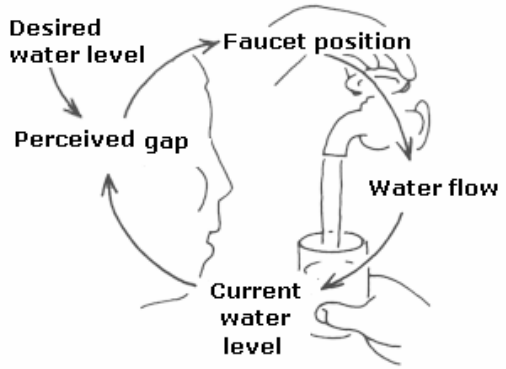


Figure 2.1: Filling a glass of water problem [41]

After understanding the structure of the problem, it is necessary to translate it into a causal loop diagram, which is a graphical representation used in SD. Causal loop dia-

grams aid visualization of how interrelated variables affect one another. These diagram consists of a set of nodes representing the variables connected together [46]. The desired water level, the faucet position, the water flow, the current water level and the gap as variables are all depicted. The gap variable is the difference between the desired level and the current level.

The relationships between these variables, represented by arrows, can be labelled as positive or negative. An arrow (causal link) from one element A to another element B is positive either if A adds to B or if a change in A produces a change in B in the same direction (i.e. if A increases, B increases; if A decreases, B decreases.) [41]. For example, in our case, if the water flow increases, the current water level will increase. The causal link between A and B, on the other hand, is negative either if A subtracts from B or a change in A produces a change in B in the opposite direction (if the current water level increases, the gap between the current level and desired level will decrease) [41].

The causal loop diagram for our example is shown in Figure 2.2. In the centre of the diagram there is a sign that identifies if the loop is positive or negative. A loop is positive when the number of negative links is even. Otherwise, the loop is negative, as in our example:

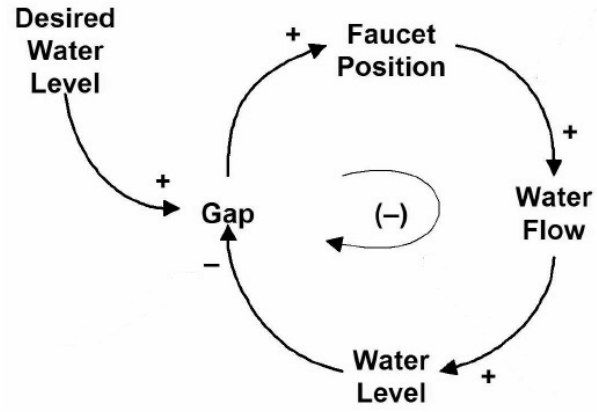


Figure 2.2: Filling a glass of water problem: Causal loop diagram [41]

A problem can further be represented by stock and flow diagrams [46]. These also show the relationships among the variables which have the potential to change over time.

The difference between these two types of graphical representations is that, unlike the causal loop diagram, the stock and flow diagram distinguishes between different types of variables, i.e. stocks, flows, information, auxiliaries and parameters.

Stocks (also known as levels, accumulations or state variables) accumulate some value over time. In our example, a stock would be the water level. Stocks are represented in the diagram as boxes. Their values change over time by accumulating or integrating flows (also known as rates, activities or movements) that represent the movement of something from or to a stock. Flows are represented by hourglasses and arrows that indicate if they are inflows or outflows from stocks. Information (curved arrows) between stocks and flows indicates that an information about a stock influences a flow. Auxiliary variables are represented by a circle and are used when one or more intermediate calculations are needed. Furthermore, they can be used as parameters in the system.

In our example, the current water level would be a stock, the water flow would be an inflow, the gap and faucet position would be auxiliaries and the desired water level would be a parameter. The stock and flow diagram is shown in Figure 2.3. In the figure, the stock is represented by the box , the flow variable is represented by the hourglass , flow , auxiliary variables , parameters and information .

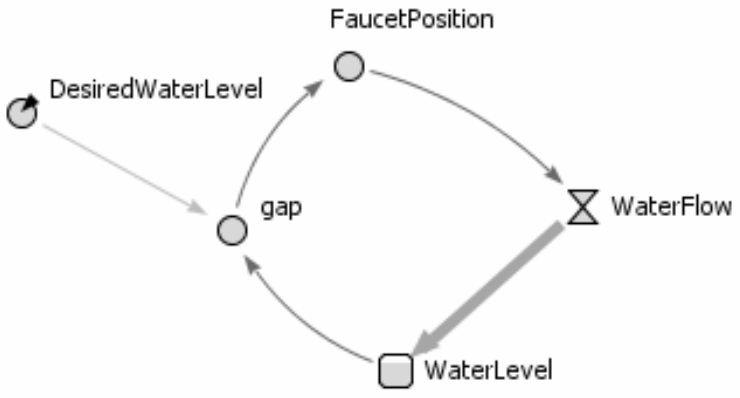


Figure 2.3: Filling a glass of water problem: Stock and flow diagram

System Dynamics Simulation (SDS) is a continuous simulation for an SD model. It

consists of a set of ordinary differential equations that are solved for a certain time interval [52]. If we consider, therefore, the stock and flow diagram of Figure 2.3 and the differential equation shown in Equation 2.1, we would have a simulation output (for 20 time units) as shown in Figure 2.4.

$$\frac{dCurrentWaterLevel}{dt} = WaterFlow \tag{2.1}$$

where:

- $DesiredWaterLevel = 10$,
- $gap = DesiredWaterLevel - WaterLevel$,
- $FaucetPosition = \frac{gap}{3}$,
- $WaterFlow = FaucetPosition$.

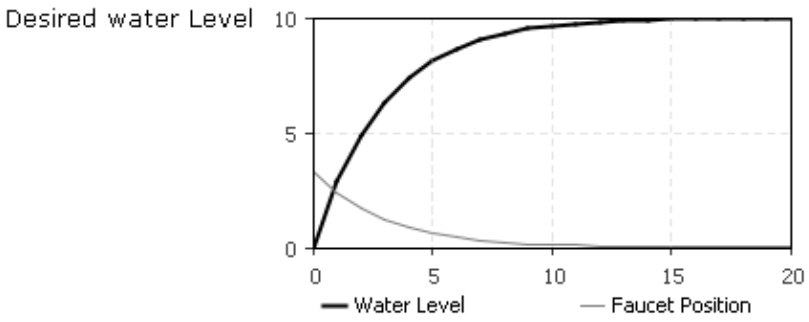


Figure 2.4: Filling a glass of water problem: SDS

2.3.2 Agent-based Modelling and Simulation

In this section, we introduce the basic concepts concerned with agent-based modelling and simulation. We start by presenting one of the first approaches to implementing agent-based models, i.e. cellular automata (CA).

Cellular Automata (CA)

CA is a discrete model consisting of two main components. The first component is an infinite regular grid of cells, which constitutes the universe or space of the CA [60]. In computer simulations, however, due to space limitations, the CA space is predetermined and finite. The second component is a finite automaton (or cell). Each cell from the grid contains a finite number of states and a predefined set of cells called *neighbourhood*. Figure 2.5 shows examples of classical neighbourhoods defined by Von Neumann and Moore. The communication of a cell with other cells within its neighbourhood is local, deterministic, uniform and synchronous [92].

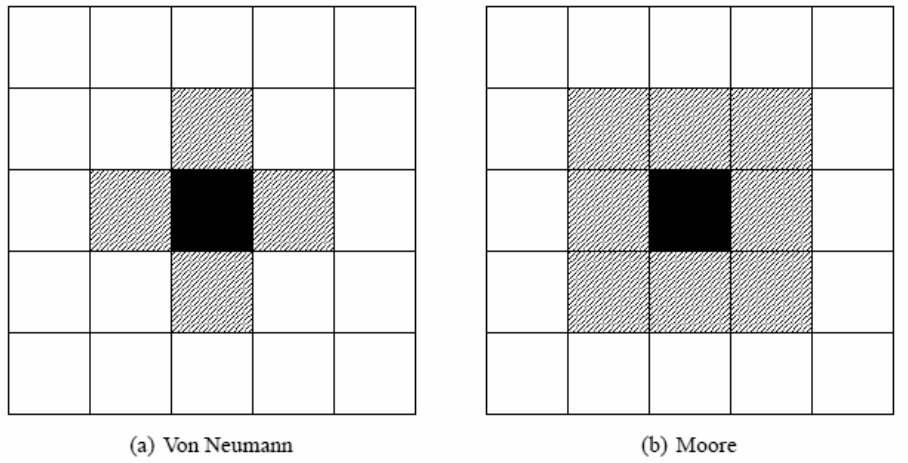


Figure 2.5: Examples of classical neighbourhoods in a two-dimensional grid CA

Each cell is initialized with an initial state at time $t = 0$. As time advances, the cells are updated according to a fixed rule, which is, in general, a mathematical function. This rule defines the next state of each cell according to its current state and its neighbourhood states.

It is possible to simulate real or abstract systems with spatial extent using CAs. They are a very useful modelling platform, as cells can represent elements of dynamic phenomena, such as individuals, attitudes or actions. CA models any world in which space can be represented as a uniform grid, elements interact with their neighbourhood, time advances

by steps, and the "laws" of that world are represented by a uniform set of rules.

Although CAs are also considered agent-based simulation by some researchers, the individuals in CA do not have memory and their interactions are limited to the individuals within their neighbourhood. In ABMS, on the other hand, agents have memory and have the capacity to interact with any other agent in the system, according to the rules established [79].

ABMS is a modelling and simulation tool, the design of which is based on artificial intelligence using concepts of robotics, multi-agent systems and complex adaptive systems [53]. It therefore employs a set of autonomous agents that interact with each other in a certain environment [93]. As it is derived from complex systems, its baseline is the notion that systems are built in a bottom-up perspective. In other words, an understanding of the system dynamics arises from individual interactions and their environment [54].

The agents' behaviours are described by rules that determine how they learn, interact with each other and adapt. The overall system behaviour is given by the agents' individual behaviours as well as by their interactions. ABMS is therefore well suited to modelling and simulating systems with heterogenous, autonomous and pro-active actors, such as human-centred systems, biological systems, businesses and organizations [79].

Another important feature of ABMS is its natural representation of a system [79]. It is possible, therefore, to mimic the real-world system by modelling entities and behaviours intuitively. The information-gathering concerned with the problem description should provide the agent-based modeller with the capacity of determine the agents' elements (attributes, associated rules, reactive and proactive behaviours).

Agent

Although there is no consensus about a definition of an agent among the ABMS community [79], Macal [52–54] defines some characteristics (Figure 2.6). An agent is:

1. A self-contained, modular, and uniquely identifiable individual. An agent has a set of attributes, the values of which will define it as an unique individual in the system.

2. Situated in an environment where the interactions with other agents occur. An agent has the capability to respond to the environment and has protocols to communicate with other agents. Its responses to environmental stimulus and interactions is defined by rules that determine the agents reactive and proactive behaviours. Apart from behavioural rules, agents communicate with each other through message exchange.

3. Autonomous and self-directed.

4. Flexible, with the ability to learn and adapt its behaviours according to the environment and past experiences (it also has memory).

5. Goal-directed, having objectives to achieve determined by its behaviour.

6. A construct with states that varies over time.

7. Social, having dynamic interactions with other agents that impact on its behaviour.

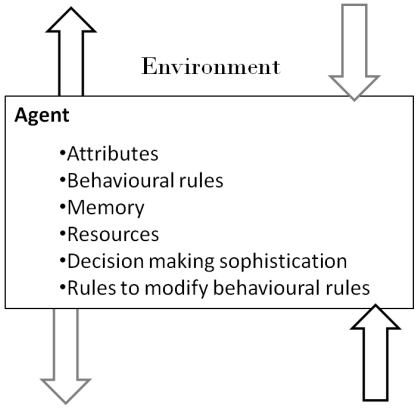


Figure 2.6: An agent (obtained from [53])

Agent-based Modelling Technique

Agent-based modelling (ABM) can be done using state chart diagrams from the unified modelling language (UML). With state charts it is possible to define and visualize the agents' states, transitions between the states, events that trigger transitions, timing and agent actions [8].

In order to understand how the state chart diagram represents an agent, let us consider an example of dendritic cell dynamics [35]. Dendritic cells are immune cells that search for signs of danger (possible antigen) in the organism [56]. They are initially in an immature state until they come in contact with a possible antigen, where they become activated. After activation, the dendritic cell investigates if it is a real danger or not. If it is a real pathogen, the dendritic cell becomes mature; otherwise, it goes to a semi-mature state. The mature state indicates that an immune response should be triggered and information about the antigen should be kept as history. The semi-mature state indicates that the foreign material should be tolerated by the immune system. Dendritic cells in an active state have a lifespan of days, while immature cells exist for longer. For our example, we consider ten days for active cells and thirty days for immature cells. After the life time elapses, cells go to a final state, i.e. death. The state chart modelling this system is shown in Figure 2.7.

2.4 Comparing Simulation Approaches

As we mentioned before, the SD is implemented by using ODEs. Their differences are therefore in the way the system is modelled, although the results in general should be the same.

The differences between SD and ABMS start with the problem representation. SD uses stock, flows and feedback loops for modelling. Moreover, SD is a continuous approach while ABMS can be either continuous or discrete. Furthermore, SD, represents entities at an aggregate level and in continuous quantities [84]. ABMS, on the other hand,

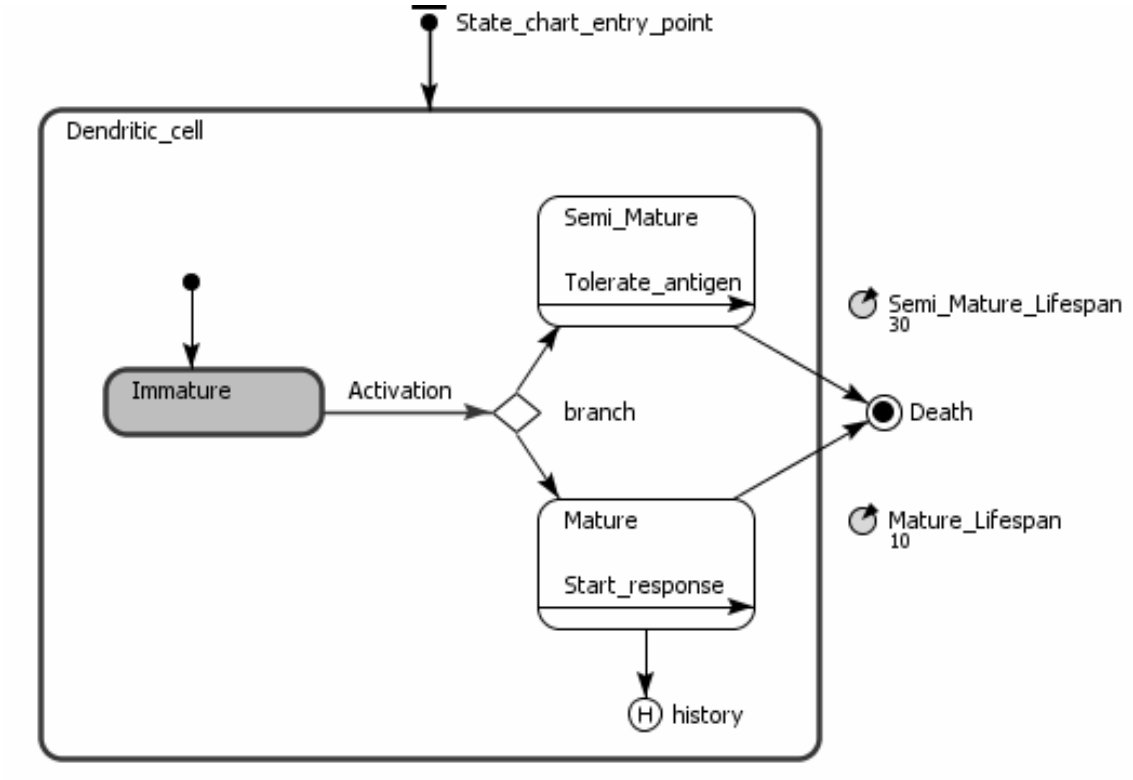


Figure 2.7: Dendritic cell state chart. The round squares represent the states and the arrows represent the transitions between the states. Arrows inside states indicate the agent actions (or behaviours).

represent individual entities that can be tracked through the system, however, only ABMS entities are capable of keeping a record of past activities (memory). SD is a top-down approaches while ABMS is a bottom-up approach.

Schieritz and Milling [76] study the differences between SD and ABMS and contrast the primary conceptual predispositions underlying each approach (Table 2.1).

SD is widely applicable at a high level of abstraction (further explanation about levels of abstraction is given in Section 2.4). ABMS, on the other hand, is a paradigm that can be used at any level of abstraction, including those levels covered by SD. As there is an intersection, a range of simulation problems can be solved either by SD or ABMS. Thorne *et al.* [86] state that ABMS is ideal for tissue patterning events because it explicitly represents individual cells in space and time. Moreover, ABMS indicates how

Table 2.1: Main differences between SD and ABMS (obtained from [76])

Feature	SD	ABMS
Perspective	top-down	bottom-up
Building block	feedback loop	agent
Unit of analysis	system structure	agent's rules
Level of modelling	aggregate	individual
System structure	fixed	not fixed
Time handling	continuous	discrete

the tissue behaviour emerges from the interactions of individual cells. On the other hand, ABMS requires computational power and may produce large sets of data, which could be difficult to analyse [86]. In addition, ABMS requires all properties of a system to be modelled discretely. SD, however, deals with continuum approximations. For the simulation of biological systems, both approaches are therefore useful and should be selected carefully according to the research question to be addressed. We will further discuss the comparison and suitability of these two approaches in section 2.5.

Simulation Modelling Abstraction Levels

Borschev and Filipov define abstraction level as the amount of detail to be included when modelling a system [8]. They categorize models in three levels of abstraction: high, medium and low.

A high level of abstraction encompasses models with less representation of details, the focus of which is the structure of the system. The model actors (elements) behave collectively as aggregates. SD and ABMS, therefore, are suitable for modelling systems at a high level of abstraction. A range of simulation problems, therefore, can be solved either by SD or ABMS. In this thesis, investigations were conducted considering the range of problems inside the intersection between SD and ABMS, as it will be further explained in the following sections.

A medium abstraction level, i.e. a tactical level, involves models with complex operational patterns, activity and decision-making. ABMS is a possible approach for this

modelling abstraction level.

A low abstraction level (operational level) involves more details, individual interactions and heterogeneous behaviours [53]. ABMS is used at this level, although ABMS models a wider range of problems at a low abstraction level.

2.5 Related Work

In this section, we describe the literature concerned with the comparison between SD and ABMS for different simulation domains. We start our review by showing general work that has been carried out to assess the differences of both approaches. Subsequently, we focus on research concerned with the comparison of the strategies for immunological problems. We found that there is a scarcity of literature comparing the two approaches for immune simulation. We also review the work related to the conversion of simulation paradigms, as the establishment of a framework for the translation between SD and ABMS is one of the main goal of this thesis.

2.5.1 Contrasting SD and ABMS

Scholl [78] gives an overview of SD and ABMS, describes their areas of applicability and discusses the strengths and weaknesses of each approach. The author also tries to identify areas that could benefit from the use of both methodologies in multi-paradigm simulations and concludes that there is little literature concerned with the comparison of both methodologies and their cross studies.

Pourdehnad *et al.* [65] compare the approaches conceptually by discussing the potential synergy between them to solve problems of teaching decision-making processes. The authors explore the conceptual frameworks for SD and ABMS to model group learning. In addition, they show the differences between the approaches in order to propose their use in a complementary way. Furthermore, they mention the lack of knowledge in multi-paradigm simulation involving SD and ABMS.

Stemate *et al.* [82] also compare these modelling approaches and identify a list of likely opportunities for cross-fertilization. The authors argue that this list should be a starting point for other researchers to take such synergistic views even further.

Schieritz [73] and Scheritz *et al.* [76] present a cross-study of SD and ABMS. They define their features and characteristics and contrast the two methods. In addition, they suggest ideas of how to integrate both approaches. Continuing their studies, [75] they then describe an approach to integrate SD and ABMS for supply chain management problems. Results show that the integration of SD and ABMS does not produce the same outcomes as SD simulation alone. To understand why these differences occur, the authors propose new tests as future work.

Demirel [19] compares SD and ABMS as models of a supply chain system. The author shows that although there are factors and effects captured by SD, it misses the dynamics at a more detailed level. These details result from the emerging heterogeneity among individual agent behaviours. Moreover, there were cases in which SD did not capture the dynamics produced by the ABMS even at an aggregate level. This happened because the SD approach does not differentiate among individuals. Any emergent behaviour that arises from individual interactions, will therefore not be captured by SD.

Ramandad *et al.* [67] compare the dynamics of a stochastic ABMS with those of the analogous deterministic compartment differential equation model for contagious disease spread. The authors convert the ABMS into a differential equation model and examine the impact of individual heterogeneity and different network topologies. The deterministic model yields a single trajectory for each parameter set, while stochastic models yield a distribution of outcomes. Moreover, the differential equation model and ABMS dynamics differ in several metrics relevant to public health. The response of the models to policies can also differ when the base case behaviour is similar. Under some conditions, however, the differences in means are small, compared to variability caused by stochastic events, parameter uncertainty and model boundary.

Schieritz [74] analyses two arguments given in literature to explain the superiority of

ABMS compared with SD: (1) "*the inability of SD models to explain emergent phenomena*" and (2) "*their flaw of not considering individual diversity*". In analysing these arguments, the author considers different concepts involving simulation research in sociology. Moreover, the study identifies the theories of emergentism that underlie the SD and ABMS approaches. The author points out that "*the agent-based approach models social phenomena by modelling individuals and interactions on a lower level, which makes it implicitly taking up an individualist position of emergence* [72]; *SD, on the other hand, without explicitly referring to the concept of emergence, has a collectivist viewpoint of emergentism, as it tends to model social phenomena on an aggregate system level*".

As a second part of the study, the author compares SD and ABMS for modelling species competing for resources to analyse the effects of evolution on population dynamics. The conclusion is that when individual diversity is considered, it limits the applicability of the SD model. However, it is shown that "*a highly aggregate more SD-like model of an evolutionary process displays similar results to the ABMS*".

Similarly, Lorenz [49] proposes that three aspects be compared and that this helps with the choice between SD and ABMS: structure, behaviour and emergence. Structure is related to how the model is built. The structure of a model in SD is static, whereas in ABMS it is dynamic. In SD, all the elements, individuals and interactions of the simulation are developed in advance. In ABMS, on the other hand, agents are created or destroyed and interactions are defined through the course of the simulation run. The second aspect (behaviour) focuses on the central generators of behaviours in the model. For SD the behaviour generators are feedback and accumulations, while for ABMS they are micro-macro-micro feedback and interaction of the systems elements. Both methodologies incorporate feedback. ABMS, however, has feedback in more than one level of modelling. The third aspect lies in their capacity to capture emergence, which differs between the two methodologies. The author states that ABMS is capable of capturing emergence, while the one-level structure of SD is insufficient in that respect. This statement by the authors about emergence differs from those previously presented

by [74].

Wayne *et al.*[89] applies SD and ABMS to simulate non-equilibrium cellular ligand-receptor dynamics over a broad range of concentrations. They concluded that both approaches are powerful tools and are also complementary. In their case study, they did not indicate a preferred paradigm, although they state that, intuitively, SD is an obvious choice when studying systems at a high level of aggregation and abstraction. SD, however, is not suitable for modelling receptors and molecules and their individual interactions.

2.5.2 Converting between Approaches

Borshchev and Filipov [8] create a practical reference to convert from SD to ABMS and present a discussion concerned with the merits of each approach. The authors use two classic SD models (bass diffusion and predator prey) and show how to convert the stock and flow diagram into state charts. In addition, they present guidance on how to convert constant and proportional rates, multiple stocks and compositionality from the stock and flow diagram to the correspondent delays in the state-charts of asyncronous agents (Figures 2.8 and 2.9). In Figure 2.8, the authors present two cases involving the conversion of simple stock and flow diagrams to the correspondent state charts, considering constant (case A) and proportional (case B) rates for the flows.

In case A there is a stock and flow diagram is constituted by a stock A and a constant outflow R. According to the authors guidelines, the stock A should be converted into the state A in the corresponding state chart. Depending on the characteristics of the problem addressed, the agent can be either deterministic or probabilistic. In the deterministic agent, the transition delay is equivalent to the total number of agents in the state A divided by the constant R, in order to achieve similar decrease as shown in the middle graph. In the probabilistic agent the delay is given by the uniform probability density distribution (PDF) bounded by the values 0 and $\frac{A_0}{R}$.

For case B, as the rate R is proportional to the value of the stock A, the corresponding

delay of the state chart transition is given by an exponential probability density function of the variable C, as shown in the middle graph. In cases where the rates change at a discrete point in time, the delays need to be recalculated for the new rate value, as shown in the bottom of Figure 2.8.

Figure 2.9 depicts another two cases and shows the conversion from SD to ABMS. In case C on top there is an inflow R to the stock B equals to $C * B$. In the corresponding state chart, a state B was created with a transition coming from and to B to represent the increase in the agents number. Similarly to case B of Figure 2.8, the delay is an exponential probability function of the value C. In addition, after the delay occurs, the action associated with the transition is the creation of a new agent. On the second part of case C, the A stock content flows to the B stock in a rate $R = C * B$. In the conversion to the state chart, therefore, there will be two states A and B with a transition from A to B with an exponential delay based on C. Once the transition occurs, the action associated is that one agent in the current state A should transit to the state B.

Case D addresses external and internal stocks influencing the flow values. The stock and flow diagram of external influence considers a stock G which value affects the outflow R. In the corresponding ABMS there is the need of two different state charts, one representing G and another equivalent the A flow. Similarly to case C (forced conversion), there is a loop transition (with delay equal to $exponential(C)$) in the state G that determines the change of state of the agents in A. Regarding the internal influence, the stock A will influence itself and, according to the Borshchev and Filipov [8] guidelines, agents at any state become B.

Finally, at the bottom of Figure 2.9 the authors show how to implement compositionality, where each constant or function will have a corresponding transition in the stock and flow diagram.

Macal [52] shows how to translate a SD into an equivalent time-stepped, stochastic ABMS. Probabilistic elements in the SD model were identified and translated into probabilities that were used explicitly in the ABMS model. The author uses as an example

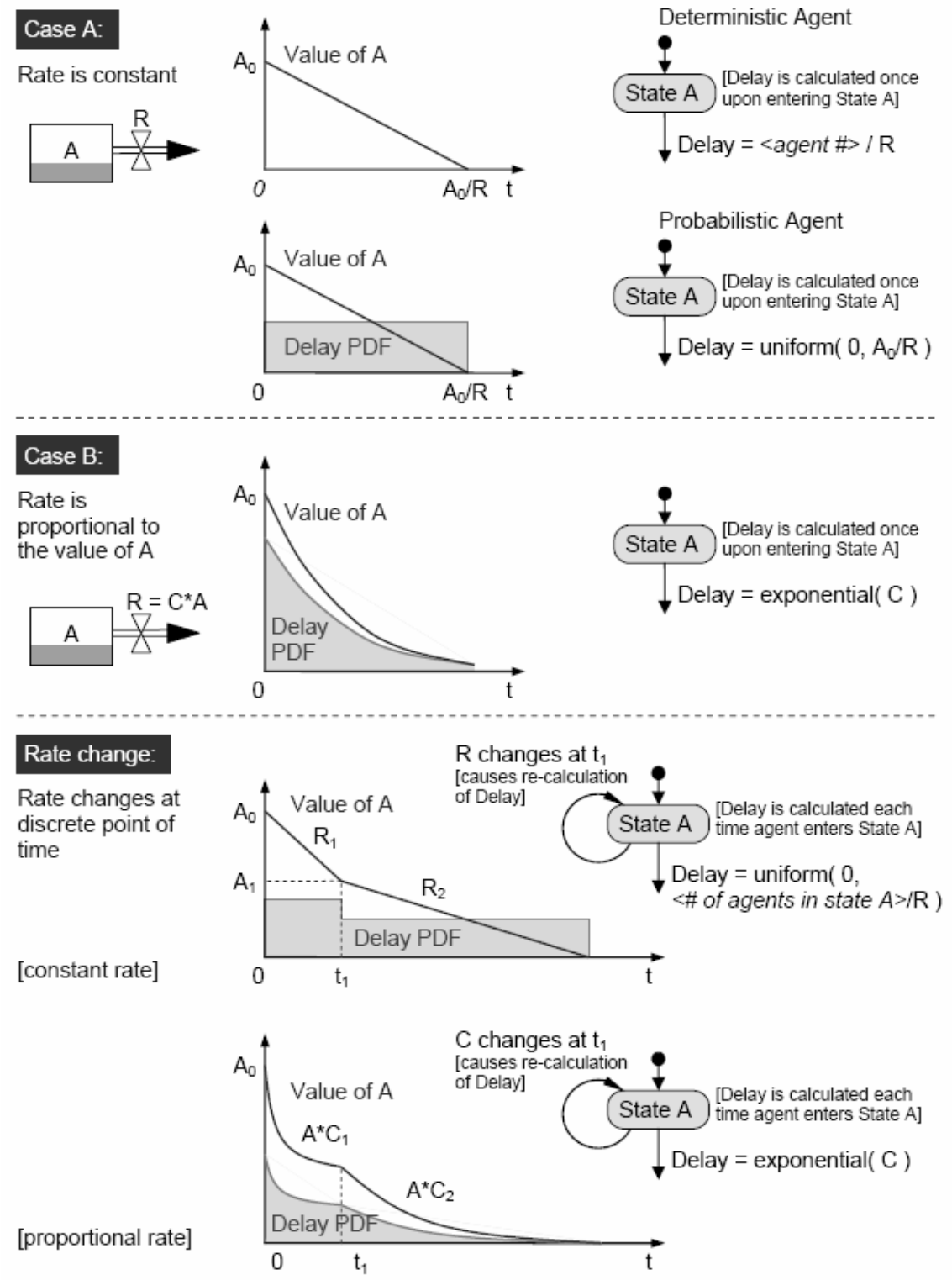


Figure 2.8: Correspondence of SD and ABMS: Constant and Proportional rates (obtained from [8]).

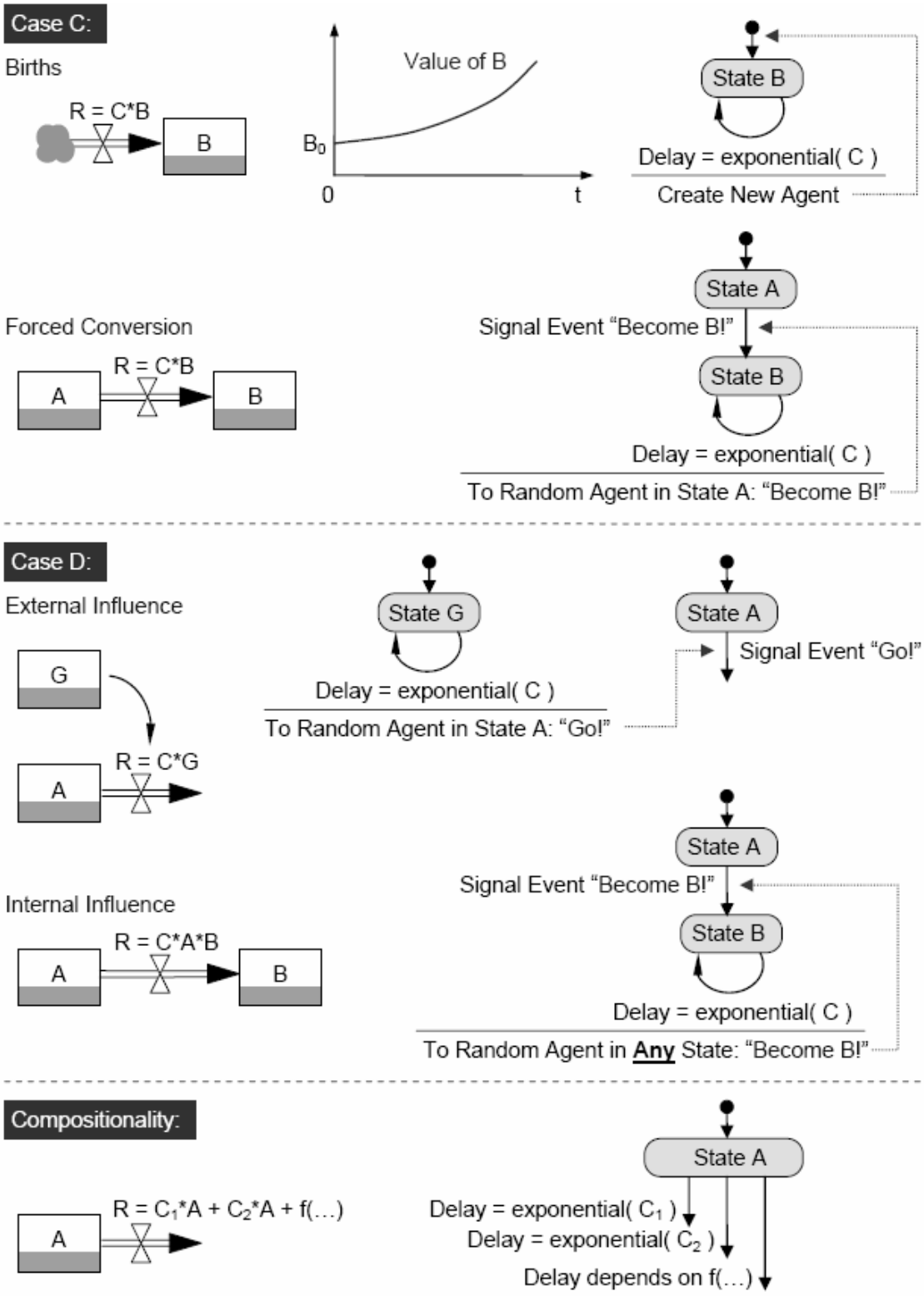


Figure 2.9: Correspondence of SD and ABMS: Multiple Stocks and Compositionality (obtained from [8]).

the SIR model proposed by Kermack and McKendrick [40]. This model was built to understand and predict the spread of epidemics. In the model, the population is divided in three groups of individuals: susceptible (S), infected (I) and recovered (R). To convert the model from SD to ABMS, the author considers two agent-based formulations. Model 1 is defined as a *"naive ABMS model, because it provides no additional information or implementation advantages over the SD model"*. There is a set of agents containing a state (S, I or R), which is the only information dynamically updated. The author claims that Model 1 produces exactly the same results for the numbers of S, I and R over time as does the SD model for a fixed-time step, Δt of length one. Model 2, on the other hand, is a fully individual-based agent model and provides additional information over the SD model. For example, as in some of the ABMS simulation runs of Model 2, an epidemic does not occur. In a significant number of cases, the number of contacts and the number of infected individuals (I) is not large enough to spread the infection. These cases occur because of the agent's probabilistic rules. Hence, Model 2 presents similar results from SD, but these are not exactly the same because of the runs where there was not an epidemic. The author therefore concludes that the ABMS model is able to provide additional information over that provided by the SD model, given its stochastic nature.

2.6 The Immune System

Life is based on a fragile balance of millions of simultaneous biochemical reactions and events, such as the interactions of a living being with his biome. A common accident, like a small splinter that penetrates the skin of an individual, creates instability in the body's homeostasis. In this case, there are two possibilities: either the aggression is recognized together with the best way to fight against it, or the individual succumbs. The immune system is the organism's apparatus of recognition, action and control towards biochemical harms [37].

If a piece of live tissue that has been extracted from a certain organism is inserted in another individual from another species, in a few days this tissue will be destroyed and eliminated by the receptor's immune system [87]. This process, known as the immune response, is characterized by the organism's ability to identify and eliminate foreign material, without implying physiological or pathological consequences of this reaction.

The immune system has the means to adapt in order to protect itself from pathogens that penetrate the organism during the life of an individual. When pathogens enter the organism, cells and molecules are produced to fight them. There are also innate defense mechanisms, which have evolved within a species and which provide a broad but not discriminated front of defense [13].

The union of the immune system's cells, molecules and tissues constitutes the mammal's global defence mechanism against antigens. This mechanism has an architecture with multiple layers and defence elements spread all over its domain [87]. An antigen is any molecule that can specifically bind to an antibody [87]. To be considered antigenic, molecules must be rigid and chemically complex. Hence, macromolecules such as proteins are examples of antigens. Among the antigen molecule surface, there are many binding sites which are named antigenic regions. The substances that make up part of the antigenic regions must be non-tolerated by the individual's immune system. Furthermore, a substance is antigenic when it is not recognised by the immune system

as self.

The first immune layer of defence against antigens – that is, the physical barriers – is composed of organs and systems which are supposed to have the first contact with the source of harm, i.e. the antigen. The main function of the physical barriers is to prevent the antigen from penetrating the interior of the organism. It constitutes the skin, which works as a shelter against any invasion, the respiratory system, which apprehends any particle through the hair, nasal mucous and activation of mechanisms such as cough and sneeze, and the mucous membranes of the digestive system.

Furthermore, it is possible to identify the biochemical barriers such as saliva, gastric acid, tears, pH and body temperature. These barriers help to eliminate the establishment of antigens in the host by providing an unfavourable environment.

The next two layers are represented by parts of the innate and adaptive immune system working together forming different types of immune responses, which can be humoral-mediated or cellular-mediated. These types of responses together with the elements involved on them are further discussed in the next sections. Section 2.6.1 addresses the characteristics of the humoral immune responses. Section 2.6.2 introduces cellular immune responses, presenting the main cells involved in the immune reactions. Finally, Section 2.6.3 introduces a new research area in immunology which addresses the phenomena of immune system aging, also known as immunosenescence. One of the aspects of immunosenescence will be further studied in one of the simulation case studies, presented in the next chapter.

2.6.1 Humoral Immune Response

Humoral immunity is mediated by molecules found in the humours, namely antibodies. The antibodies are produced by a type of lymphocyte, known as B cell. Their production is stimulated by the presence of an antigen. The antibodies react chemically against the antigens and neutralize their harm in the organism. Antibodies are antigen specific, which means that for a certain antigen there is a set of antibodies specially produced

for its neutralization [37].

The immune response process can be understood with an example of the production of antidotes for snake poisoning. The antidote is obtained from antibodies on horses which have been infected with small quantities of poison. As their immune system starts to react against the toxic substance, the specific antibodies are extracted from the blood and used as antidotes for another animal.

There are no changes in the horse's blood serum for several days after the first exposure to poison. After this lag period, there is a high concentration of antibodies, which reaches a maximum and then decays quickly. This characterizes the primary immune response. If a new poison dose is injected sometime after the first reaction, the secondary response will occur faster (Figure 2.10). The difference between primary and secondary responses indicates that the immune system has some kind of memory [87]. Memory provides the immune system with the ability to protect itself faster when re-exposed to antigens. In certain cases, the immune memory may have its dimension reduced by time. However, it continues providing faster secondary immune responses.

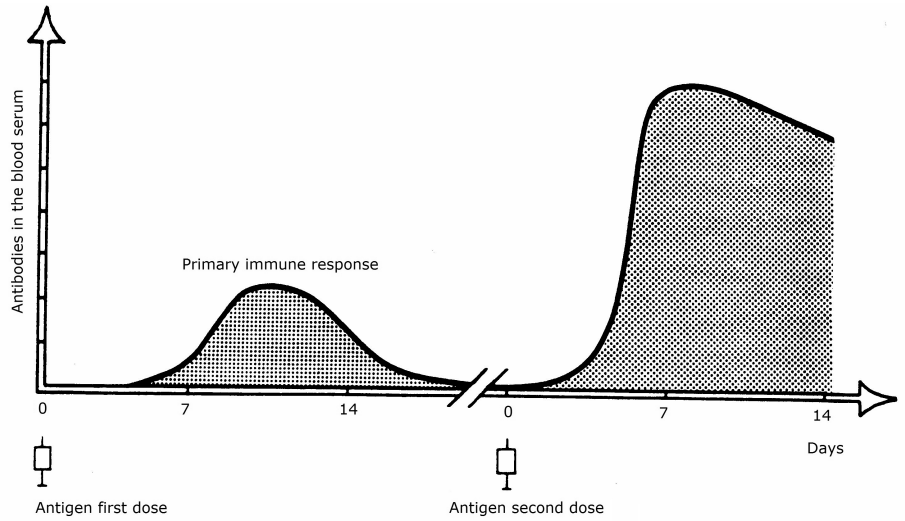


Figure 2.10: Primary and secondary immune responses (obtained from [87]).

2.6.2 Cellular Immune Responses

When a foreign tissue is inserted in an individual, in approximately ten days the organism will have completely removed this tissue. If another similar tissue happens to be inserted in the individual, the removal process will take less than two days. While this is similar to the process that occur in humoral responses (Section 2.6.1), in this case, there are also secondary and primary immune responses. For this type of immune response, however, the main actors are immune cells.

Another context in which cellular responses can be found is in the defence against intracellular parasites, such as viruses. Viruses are microorganisms composed of nucleic acid surrounded by a protein case. Because they do not have a reproductive system, they invade cells and make them work as a virus replicating system. The viral infection, therefore, affects the cell's metabolism, even causing cellular death. While inside the cell, a virus is protected from antibodies by the cellular plasmatic membrane. In order to fight these types of infections, there are immunocompetent cells specialized in searching and destroying any kind of anomalous cell in the organism.

These immunocompetent cells are the white blood cells, or leukocytes. They defend the body against both infectious disease and foreign materials. Leukocytes are found throughout the body, including the blood and lymphatic system. There are several types of white blood cells, which include different types of lymphocytes and phagocytes [37].

A lymphocyte is a type of white blood cell. There are two broad categories of lymphocytes: the large granular lymphocytes and the small lymphocytes. In general, large granular lymphocytes are natural killer cells (NK cells), while T cells and B cells are small lymphocytes.

Natural Killer Cells

NK cells are a part of the innate immune system and play a major role in defending the host from both tumours and virally infected cells. NK cells distinguish infected

cells and tumours from normal cells by recognizing alterations in a cellular surface molecule, named MHC (major histocompatibility complex) class I. NK cells are activated in response to molecules, known as cytokines. Activated NK cells release cytotoxic (cell-killing) granules which destroy the anomalous cells [37].

B Cells

Each B cell has small sites on its surface, namely paratopes, which specifically detect molecules. Paratopes are responsible for antigen recognition via antigenic determinant identification, i.e. epitopes, which are sites against which immune response tends to react. Furthermore, these are areas where the bindings of antibodies take place. A B cell becomes activated if it binds and recognizes an antigen. During the activation process, a B cell increases in size and reproduces, and the offspring differentiate into antibody secreting cells (plasmocytes) and memory cells. While replicating, B cells suffer a process called *affinity maturation*. This process allows for mutations in the B cell's paratopes, known as *somatic hypermutations*, in order to improve the affinity between antibodies and antigens [37].

Antibodies are protein molecules that binds and neutralizes an antigen (Figure 2.11). This binding occurs between the paratope of the antibody and the antigen's corresponding epitope [37].

T Cells

T cells represent most of the blood lymphocytes and are part of the adaptive immunity. As well as B cells, T cells have their origin in the bone marrow. Their maturation, however, occurs in the thymus. In the maturation stage, lymphocytes have their function and antigenic specificity defined. The most important types of T cells are T regulatory cells and cytotoxic T cells [37].

There are two subtypes of T regulatory cells: T helper cells and regulatory T cells (T_{Regs}). T helper cells are responsible for the secretion of lymphokines, which are

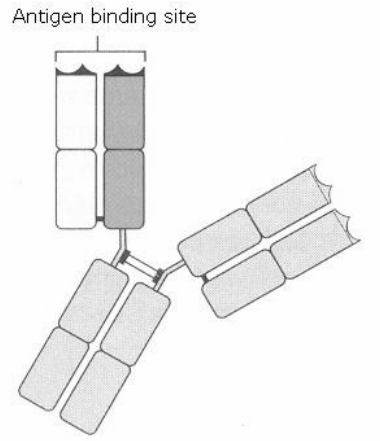


Figure 2.11: The antibody (obtained from [37]).

soluble factors similar to hormones that affect other cells. They start the activation and transformation of B cells in plasmocytes and they are also partially responsible for the activation of cytotoxic T cells and inflammatory white blood cells [13].

T_{Reg} cells inhibit immune responses and even directly suppress a certain B cell function. They are also capable of suppressing the activation of a T helper cell, being in certain cases, antigen-specific.

T cytotoxic cells play a main role in the cellular immune responses. They act by destroying the anomalous cells and those cells which are pathogen-infected.

The division of T cells into two subclasses was made possible after the discovery of a relation between the role played by a certain T cell and some membrane-expressed proteins. Most T helper cells express a CD4 (cluster differentiation 4) surface protein. The expressed protein in cytotoxic T cells is CD8. Following these patterns, it was possible to identify many types of B and T cells. The cluster differentiation (CD) is a protocol used for the identification and investigation of cell surface molecules present on leukocytes. CD molecules can act in numerous ways, which often are receptors or ligands (the molecule that activates a receptor) that are important to the cell [13].

A third group of cells was identified as not having membrane markers with specific characteristics that would identify them as B or T cells. In this group we can find the

natural killer cells.

Phagocytes

Phagocytes are cells found in the blood, bone marrow and other tissues of vertebrates. Phagocytes ingest antigenic and infectious agents in the body. They originate in the bone marrow and are the basis of defence in the innate immune system; these cells ingest pathogens in a process called phagocytosis and often take part in antigen presentation. The types of phagocytes include macrophages, neutrophils and dendritic cells [37] (Figure 2.12).

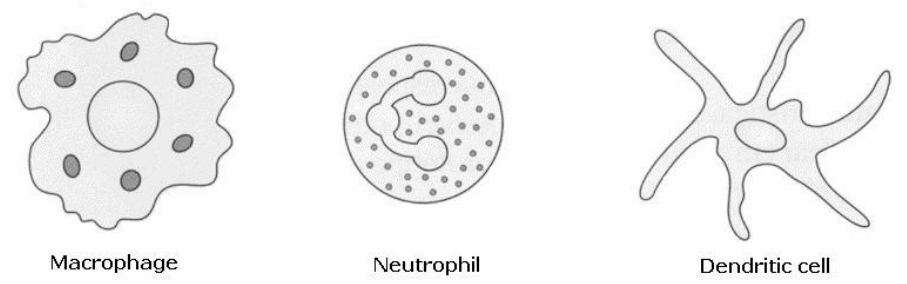


Figure 2.12: Phagocytes (obtained from [37]).

Macrophages are widely distributed in the blood and tissues. In their immature form, they are known as monocytes. When a monocyte enters damaged tissue through the endothelium of a blood vessel, it undergoes a series of changes to become a macrophage. Monocytes are attracted to a damaged site by chemical substances through chemotaxis, triggered by a range of stimuli including damaged cells, pathogens and cytokines released by macrophages already at the site [34].

Macrophages survive in the body up to a maximum of several months. Among the important roles of the macrophage are the removal of necrotic cells and the presentation of antigens. After digesting a pathogen, a macrophage will present the antigen of the pathogen to the corresponding helper T cell.

Granulocytes increase during immune responses and migrate from blood to infection or inflammation sites. Neutrophil granulocytes, generally referred to as neutrophils, are

the most abundant type of white blood cells in humans and form an essential part of the immune system. Being highly motile, neutrophils quickly congregate at a focus of infection, attracted by cytokines.

The function of dendritic cells is to catch and present the antigen to the lymphocytes, so that the pathogen can be eliminated. They have surface receptors capable of recognizing the common structures of a wide range of pathogens. As these receptors find a pathogenic molecule, the dendritic cell is stimulated to englobe the pathogen and degrade it into peptides. Thereafter, the dendritic cell becomes activated and migrates to the nearest lymph node to present these peptides to the specific lymphocyte, which will determine how the immune system must respond to the antigen [34].

2.6.3 Immunosenescence

Ageing is a complex process that negatively impacts on the development of the immune system and its ability to function [10]. Progressive changes of the components of immune systems have a major impact on the capacity of an individual to produce effective immune responses[61].

The decrease of immunocompetence in the elderly can be envisaged as the result of the continuous challenge of the unavoidable exposure to a variety of potential antigens, e.g. viruses [62], bacteria, food and self-antigens [33]. Antigens are the cause of persistent life-long antigenic stress, responsible for the filling of the immunological space by an accumulation of effector cells and immunological memory [55].

With age, there is also a significant reduction in the number of naive T cells caused by the involution of the thymus [20]. Naive T cells are responsible for responding to new faced antigens. The reduction of these cells eventually leaves the body more susceptible to infectious and non-infectious diseases [59]. There is also evidence that clones of immune cells deteriorate [11, 57], while innate immunity is conserved or even up-regulated [33, 94].

In addition, the ageing of the immune system is responsible for the pathogenesis of

chronic age-related diseases such as arthritis, atherosclerosis, osteoporosis and diabetes [10, 44]. With age, there is the up-regulation of the inflammatory responses, due to the chronic antigenic stress that impinges throughout life upon immunity [62], and has potential implications for the onset of inflammatory diseases. The chronic exposure to stress factors also leads to a progressively reduced capacity to recover from stress-induced modifications and decrease in vaccine responsiveness.

2.7 The Need for Simulation of the Immune System

An important issue raised by Ulgen *et al.* [88] concerned with simulation for industry is to determine whether simulation is actually needed or whether mathematical models are sufficient. We believe that this issue should also be considered for immune system modelling. In addition, for immunology (and biology in general), there is another competing approach, which is based on methodological reductionism [71]. This approach investigates biological phenomena by looking at its molecular and cellular levels, disregarding emergence [9].

In many studies, models such as spreadsheet analysis, mathematical programming and optimization approaches (such as linear programming and branch and bound technique) or statistical modelling techniques (such as regression modelling) are more appropriate to use than simulation [88]. For example, Murray *et. al* [59] introduce a regression model of immune cells output from thymus that fits the original data better than the agent-based the simulation approach [26]. Furthermore, Ulgen *et al.* [88] argue that the appropriateness of a mathematical model instead of simulation may become evident along the development of the simulation model. As we mentioned before, some important advances in immunology were facilitated by the joint work of immunologists and mathematicians. Many of the major concepts existing in theoretical immunology are the result of these models.

Most existing models in immunology are based on sets of differential equations [21,

36, 91]. This approach for immunology, however, limits the modelling effort to simple dynamics involving few immune elements such as cells or molecules and it only allows analysis at an aggregate level. Moreover, it is not trivial to model problems involving individual localisation, memory and emerging properties mathematically [8, 51]. Hence, simulation serves as a complement of mathematics to solve high complexity problems.

The goal of simulation is to mimic the dynamic behaviour of a real system. The major benefits of simulation to immunology, compared to mathematical models, are the capacity to model dynamic complex systems and emergence. In addition, with simulation it is possible to observe how the system evolves over time, which provides insights about the dynamics of the system, instead of just predicting the system output given a certain set of inputs [79]. Moreover, the simulation modelling methods are closer to the natural description of the system, without the need of an in depth understanding of mathematics [7].

A work presented by Sauro *et al.* [71] debates the usefulness of simulation in contrast to reductionism in biology. According to streng [22] in reductionism *"the dynamics of any complex system can be understood from studying the properties of its parts. Complex systems are therefore broken down into their components and each piece is studied individually by way of disciplinary and sub-disciplinary approaches. The challenge is to find the entry points from where to address the particulars of the system. Once one knows the parts, the dynamics of the whole can be derived"*. On the other hand, the opposite of reductionism, i.e. holism (in its methodological version) is *"the relationship between the parts and the whole is believed to be more symmetric than in reductionism. The assumption underpinning this approach is that the properties of the parts contribute to our understanding of the whole, but the properties can only be fully understood through the dynamics of the whole. The research focus in holism is on the relationships between the components, i.e. on their interconnectedness, interdependencies and interactions. In holism, the whole is more than or different from the sum of its parts"*. Simulation is, therefore, based on holism.

For biology, Sauro *et al.* [71] state that "*reductionism has proven to be a highly successful strategy and has enabled us to uncover the molecular details of biological systems in unprecedented detail. In particular, light microscopy of single cell dynamics is a reality and enables a researcher to track the concentration of a small number of proteins in real-time. Using light microscopy and cell counting techniques, large amounts of high resolution data on a small number of observations can be collected*" [71].

The success of reductionist methods raised some scepticism as to the need for alternative approaches, such as systems biology. The challenge for simulation is, thereby, to generate novel insights that can not be uncovered just by looking at a phenomena using reductionism. Examples of successful simulation approaches that helped advance immunological research were introduced in [23, 86]. The models reviewed simulate interactions of immune cells and chemical substances, humoral responses, and drug testing. With the simulations it was possible to observe emergent behaviour in the systems, which is not contemplated by reductionism.

The major benefits of simulation to immunology, compared with real-world experimentation, include time and cost effectiveness due to the labourious and resource-intensiveness of the biological environment. Most experiments are expensive and have to be in agreement with ethical specifications. Moreover, the accuracy of the results of laboratorial experiments relies on environmental conditions, the quality of the material collected and the appropriate conduct of experimental tests. For instance, blood samples can be compromised by inappropriate storage or cells can die of excess exposure to a certain chemical agent. Furthermore, in a simulation environment, it is possible to systematically generate different scenarios and conduct experiments.

Case studies from immunology research will be used along this thesis in order to evaluate our framework of conversion between approaches. In addition, we want to show the potential contribution of SD and ABMS to advance immunological studies.

2.8 Summary

Simulation techniques are powerful decision-support tools, which allow us to mimic the real-world in order to investigate how system elements progress over time. Furthermore, these techniques offer the means to understand and control a system model, test different scenarios and provide further insights about processes. The main system modelling and simulation approaches used in this thesis are system dynamics (SD) and agent-based modelling and simulation (ABMS).

SD is a continuous top-down modelling and simulation approach, based on systems theory. It uses stocks, flows and feedback loops as concepts to study the behaviour of complex systems. SD models are graphically represented by causal loop or stock and flow diagrams. In addition, SD simulation consists of a set of difference equations that are solved for a certain time interval. Hence, these models are capable of encompassing most of the mathematical simulations models.

ABMS is a bottom-up technique that employs autonomous agents that interact with each other. The agents' behaviour is described by rules that determine how they learn, interact with each other and adapt. The overall system behaviour arises from the agents' individual dynamics and their interactions.

An important point when choosing a modelling method is the required level of abstraction. It specifies the level of detail to be included in the modelling system [8]. SD is widely applicable at a high level of abstraction. Agent-based modelling, on the other hand, is a paradigm used at any level of abstraction. A range of simulation problems, therefore, can be solved either by both approaches.

ABMS explicitly represents individuals in space and time. Moreover, this approach indicates how the system behaviour emerges from the interactions of individual elements. ABMS, however, requires more computational power than SD simulations and may produce large sets of data, which could be difficult to analyse. In addition, ABMS requires all system's properties to be modelled discretely. SD, however, deals with

continuum approximations.

Sauro *et al.* [71] states that although ABMS is very useful for simulations in areas such as systems biology, there are circumstances where they cannot be applied. For instance, when the reaction network process is not well understood, or experiments are known not to be able to reproduce the real-world reactions (given the environmental differences such as temperature, for example). Furthermore, the authors argue that "*top-down modelling strategies are closer to the spirit of systems biology exactly because they make use of systems-level data*", and thereby they conclude that there is no best approach as it is preferable to view them as complementary.

With the advent of multi-scale and multi-paradigm simulation, an indepth understanding of the outcome differences of distinct paradigms and how to translate from one approach to another is imperative. However, there is little research investigating the translation between SD and ABMS.

Most existing models in immunology are based on sets of ordinary differential equations [21, 36, 91]. This approach for immunology, however, limits the modelling effort to simple dynamics involving few immune elements such as cells or molecules and it only allows analysis at an aggregate level. Moreover, it is not trivial to model problems involving individual localisation, memory and emerging properties mathematically [8, 51]. Hence, systems simulation emerged as a complement of mathematics, to deal with larger amounts of available data and high problems complexity.

These methods, however, are still not widely adopted in immunology research. In addition, to our knowledge, the processes for the development of simulation models for the immune system have not yet been formally structured. Hence, there is the need to show the importance of simulation to help immunological research and to draw the attention of simulation developers to this research field.

SD potential contribution to immunology needs to be investigated, as problem conceptualization using causal-loop/stock and flow diagrams seems to be more intuitive than the mathematical formulation. Furthermore, SD diagrams facilitate the visualiza-

tion/understanding of variables and causal effects in a problem. Immunologists might therefore find it easier to use SD as a tool for their *in silico* experimentation. For specialists familiar with numerical simulation, the transition from mathematical methods to SD is quite natural.

In the remainder of this thesis, we define a framework to convert between ODEs equations, SD and ABMS. As it is acknowledged that most simulation efforts in immunology are mathematical, we seek to develop an algorithm for the translation of mathematical models to a SD environment, as immune simulation developers would very much benefit from this tool. We also believe the conversion of SD to ABMS would be a first step towards adding emergence and individual tracking in a system model. Moreover, with the advent of research on combining both techniques, some models could benefit from a multi-paradigm (SD combined with ABMS) simulation.

The guidelines provided in Figure 2.9 are a starting point for the conversion from SD to ABMS. However, there is the need of additional investigation in order to assure their completeness. Furthermore, the application of these guidelines to theoretical case studies and real-world problems is necessary in order to explore the conversion efficiency. It is intended, therefore, to observe the outcomes of each converted approach and verify if there are any differences, and to understand why they occur. If outcomes differ, we aim to systematically test how and when these differences occur. In addition, we intend to provide insights of what should be expected when using one approach or another. As our interest lies in immune system problems, we use classical case studies of immunology found in literature.

In the next chapters, we will define our methodology (Chapter 3) and case studies (Chapters 4 and 5) considering aspects such as the behaviour of the entities of the model (whether they are static or interact with other entities and if they have spatial representation or not), the type of hypothesis to be tested, the empirical embeddedness of real data and the modelling effort.

Chapter 3

Methodology

3.1 Introduction

This chapter presents the research methodology used for the development of our simulation models and for the experimentation performed in the following chapters.

In Section 3.2 we briefly reiterate the research problem, which is addressed by the research aims and objectives devised and presented in Section 3.3.

In Section 3.4 we develop our strategy to achieve our thesis objectives. Furthermore, we introduce the steps used in the process of building immune simulations, some of which will be employed in our experiments.

As there are no explicit guidelines in literature, we have developed our own set for the conversion between approaches. These guidelines are presented in Section 3.5. In Section 3.5.1 we explain our steps to convert from ODE models to SD and, subsequently, we describe how to convert from SD to ABMS (Section 3.5.2). In Section 3.5.3 we show how to convert from ODEs to ABMS. Section 3.5.4 introduces the conversion guidelines from ABMS to SD.

Finally, in Section 3.6 we present the summary of this chapter.

3.2 Research Problem

Although there are examples of successful applications of modelling and simulation approaches to immunology, these are not commonly used by immunologists. Instead, ODE models are widely employed to support advances in major concepts in immunology. However, these models have limitations for solving complex problems which involve individual localisation, memory and emerging properties. Systems simulation is therefore a set of methodologies that has emerged to complement mathematical models and overcome some of their limitations (Section 2.7). Possible modelling and simulation approaches for immunology include system dynamics (SD)(Section 2.3.1) and agent-based modelling and simulation (ABMS) (Section 2.3.2).

Few studies apply SD to immune problems (examples can be found in [27] and [30]). ODE models are more commonly used instead. SD, however, encompasses the mathematical formulations with the advantage of retaining information of how the elements modelled in the system change over time. Furthermore, there is no need for an indepth understanding of mathematics to formulate a model. The ODEs are implicit in the system's structure and the relationships between the elements modelled can be established with experimental data (Section 2.3.1).

ABMS is more suitable for simulations in which there is the need to represent individual cells, their interactions and emergent behaviour (Sections 2.3.2 and 2.4).

The selection of a modelling and simulation technique for a problem is driven by the resources available, such as experimental data, an understanding of the mechanisms involved, the hypothesis to be tested and the level of abstraction needed to test the hypothesis. Once the system description and the simulation domain model is defined, a simulation method needs to be chosen. There is little research comparing and assessing the merits of SD and ABMS for immunology. We therefore believe that it would be useful to investigate both approaches applied to immune system problems.

As immunology is a field that constantly gathers new information, simulations have to be

updated frequently to suit new findings. For some cases, in order to meet new demands, the replacement of the current simulation approach for new developments needs to be considered. There are two options, namely, to start the new simulation from scratch or to adapt previous implementations for the new approach. However, with regard to the second option, there is little knowledge in literature on how to convert implementations between approaches and the implications of these conversions.

Another possible advantage of the conversions guidelines would be in circumstances where a model developer is well acquainted with one simulation approach and would like to use his current models to learn another technique. In cases where there is, for example, a established ODE model, conversion techniques to translate the current model to ABMS would be a good starting point to learn this simulation approach and possibly expand the model.

3.3 Research Aim, Objectives and Deliverables

Taking this research gap into account, we outline our research aims as:

- With no explicit guidelines available from the literature, we aim to develop, test and validate our own set of guidelines for converting between ODE, SD and ABMS approaches. We believe these practices will assist with the improvement and expansion of existing immune models to suit new demands.
- We seek to asses the impacts of these conversions and determine those circumstances where one approach could be replaced by another. We also want to determine, after performing the translation, when one approach would be preferable than the other.

In order to achieve our aims, the main objectives of our work are:

1. To define and test guidelines to convert between (1) ODE models to SD models,

(2) ODE models to ABMS models (3) SD models to ABMS models, (4) ABMS models to SD models and to assess the impact of these conversions.

2. To compare ODE, SD and ABMS outcomes, considering aspects such as the behaviour of the entities of the model (whether or not they are static or interact with other entities, and whether they have spatial representation or not), the type of hypothesis to be tested and the modelling effort.

3. To define guidance to choose between SD and ABMS depending on the characteristics of the problem to be addressed.

3.4 Development of a Research Strategy

In order to achieve the aims and objectives outlined above, it is necessary to define a strategic research programme to direct the activities of this research in several stages. This strategy is developed in this section following research design principles. In the next section we introduce our research strategy where the activities and methods required to realise our objectives are identified.

3.4.1 Principles of Research Design

The objectives defined in Section 3.3 suggest the type of research and hence the methodology to be used, based on the research taxonomy defined by Robinson [69] and Yin [96].

The first objective (compare SD and ABMS outcomes) suggests a deductive approach, since we are testing the hypothesis that it is possible to obtain similar outcomes for both approaches. Furthermore, it is descriptive as it determines what is actually happening, and quantitative as it needs to be able to measure absolute values.

The second objective (guidance to help immunologists to choose between SD and ABMS) also suggests a deductive approach, as we wish to test the hypothesis that it is possible to obtain similar simulation outcomes for the different simulation approaches studied.

Furthermore, the research approach will be descriptive as it provides information about the most suitable approach based on the results investigated. In addition, it is quantitative as it needs to be able to measure absolute values, such as number of cells at a certain point in time.

The third objective (define and test guidelines to convert between simulation paradigms) suggests that the approach to be used in this study should be (1) deductive, testing the hypothesis that it is possible to obtain equivalent models by following the guidelines defined; (2) exploratory, as we wish to find out if we can apply the guidelines to convert between approaches; and (3) quantitative, as we want to compare the results of the equivalent approaches by applying statistical tests.

To achieve the above objectives, we adopted a case study methodology, which is suitable and robust, given the issues faced during this research, namely time and resources constraints and problems of arranging access to experimental data and co-operation. This methodology is presented in more detail in the next section (Section 3.4.2).

3.4.2 Research Strategy

In this section we outline the activities and methods necessary to realise our objectives. We examine several case studies of established mathematical models that describe some immune mechanisms. These case studies were chosen by considering aspects such as the behaviour of the entities of the model, the type of hypothesis to be tested, the empirical embeddedness of real data and the modelling effort. With regard to the entities' behaviour, we first contrast SD and ABMS simulations when these entities are static (without any movement). Subsequently, we investigate interacting entities with and without spatial representation. Furthermore, the results are analysed and an investigation is carried out to identify which output seems more realistic in terms of real-world experimentation (for those cases where we had access to experimental data). For each aspect analysed, we use a multiple-case approach comprising of three case studies. This methodology was chosen as we believe it can provide major insights during our

experimentation stage. In addition, it allows for systematically replicating design logic and testing.

As there is no established guidance to develop simulations for the immune system, we studied those developed by [70] for simulation in operational research problems and adapted them for simulations of the immune system. The adaptation was performed by studying several simulations developed for the immune system [23, 25, 48, 86, 98]. We observed the similarities and differences with operational research and outlined general steps for building immune simulations. Furthermore, we discuss the pitfalls that might be encountered during the process, as shown below.

1. **Define the Objectives.** Overall, the objectives of simulations for the immune system are (1) to investigate a theory and/or (2) to create an environment containing "what-if" scenarios without the ethics restrictions. The scenarios can either be based on experimental data or defined as an intuition of what might happen in reality. Furthermore, there are also cases where actual models do not match real-world experimentation and they need to be further investigated (in a simulation model). In addition, new hypothesis and research questions may be defined together with immunologists as simulation goals. The objectives come from real-world observation. We assume, however, that real-world observation and experimentation has been previously performed by immunologists.

2. **Describe the system**. In this step, it is necessary to use documents (immunology books and articles, transcripts of interviews with experts, etc.) describing how the immune elements to be simulated work and interact. The description of the system is based on knowledge acquired by theoretical work, real-world observation and laboratory experimentation. Due to the complexity of the elements and processes in the immune system, however, this knowledge is scarce. The immune system is far from being fully fathomable, and the descriptions found in literature are only partial representations and assumptions of what occurs in reality.

3. **Investigate existing theories and established models.** The definition of mathematical models which describe a phenomenon observed is prevalent among immunologists. These models are generally verified using experimental data. In order to build a new simulation model, it is common to look at the existing models and investigate their hypothesis, objectives, validation process and limitations. By reviewing the existing models it is possible to build a new model as an improvement of what has already been established in order to investigate a certain immune process.

4. **Use experimental data.** Currently, most simulation models are built based on real-world experimentation. There are some models, however, where there is no data available (for example, when Jerne's network theory was conceptualized [38]). These models are based purely on theoretical assumptions with the purpose of providing more insights about what happens in the real world. Furthermore, in the field of immunology the non-existence of data can be due to the lack of understanding of a process, or a difficulty or even impossibility in collecting information with current technology.

   In other cases, a hypothesis is first formulated requiring experimental data to confirm it. There is therefore the need to collect this data. For instance, Foan *et al.* [30] implemented a SD simulation of T cell subsets throughout a person's lifetime based on an established mathematical model developed by Balcheva [3]. The authors conclude that further validation of this model is necessary and so a novel data set should be collected as there are arguably more specific markers that could help to gain further insights from the model.

5. **Build conceptual model.** The conceptual model of a problem is an abstraction intended to contain the principal aspects observed in the real world, considering the necessary level of details [43]. In this step we formally define the model scope, the objectives previously outlined, the inputs and outputs and the simplifications. The process of creating a conceptual model evolves with decisions regarding the

model scope and level of detail [70]. The acceptance of the conceptual model should be agreed with immunologists. According to Ulgen *et al.* [88], "*rigorous validation procedure for the conceptual model is as important as the verification and validation of the model because it saves time and redirects the simulation developers in the right direction before time is wasted in the study*".

Due to the limitations of immune simulation, it is important to abstract the relevant real-world features and build a simple model. According to Kotiadis and Robinson [43], the importance of model abstraction "*relies on the fact that there is no need to model all that is known about the real problem. Simpler models are developed and run faster, they are flexible, require less data and results are easier to be interpreted*". The nature of immune problems thus implies that the model should be developed in order to address a few objectives, within a limited scope. The description of the system (and definition of the conceptual model) should therefore focus on the parts of the immune system (scope, elements, information available, assumptions, hypotheses) relevant to achieve the simulation goals.

Daigle discusses the challenges of modelling immunology [18]. As it is a field in which information is still being gathered, simulations have to be updated frequently to suit new findings. Moreover, current computational resources and modelling techniques are in development. It is still thereby impossible to represent computationally an entire pool of cells of a typical immune response (around 10^{12} cells). In addition, immunological systems are mostly hierarchical, involving several layers and complex interactions between the elements of these layers.

6. **Identify elements, parameters, aggregates, etc. already established in theory and real-world data.** The study of the abstract model provides a means to understand the problem and the best way to represent the elements of the system, together with the more suitable simulation approach. For example, if the in the conceptual model it is established the interactions of the simulation will occur at a cellular population level rather than an individual cell level, this might

indicate that a top-down simulation approach would be more suitable to build the model.

7. **Decide on the most appropriate modelling and simulation approach.** This decision is made based on the characteristics of the problems, the research questions to be addressed, the scope, the level of aggregation and the experimental data available. The most common approaches used in immunology are ABMS, cellular automata and SD.

   Cellular automata is used for problems involving autonomous individual interactions within a neighbourhood and emergent behaviour. For example, individual interactions between cells and molecules, which do not involve spatial localization or memory.

   ABMS is suitable for problems involving autonomous individual behaviour, elements spacial localization, memory and emergence. For example, individual interactions between cells and molecules which demand either spatial localization or memory.

   SD defines a system at a high level of aggregation. Hence this approach should be used when the research question involves patterns of behaviours and feed-back interactions between the aggregates. This approach is very useful to simulate dynamics of populations and interactions between different populations overtime. For example, interactions between tumours and effector cells, viruses and T cells, etc.

8. **Represent elements, parameters, etc. using the appropriate modelling and simulation approach.** Once the modelling approach is chosen, the elements defined in the conceptual model need to be translated into their correspondents used by each approach, for instance, stocks, flows, parameters and information for SD or agents and rules for ABMS. This step is part of the construction of the simulation model, defined in the next step.

9. **Build the simulation model.** This stage includes the development of the computational implementation of the model in a simulation tool. The implementation is a software representation of the requisites defined in the conceptual model. The computational model is the final product to be used by the immunologists.

10. **Verify the model.** The model verification is the process of ensuring that the model design has been transformed into a computer model with sufficient accuracy [70].

11. **Validate the model with existing theories and, if available, real-world data.** Validation ensures that the model is sufficiently accurate for the purpose at hand. For immunology it is acknowledged that models are not intended to be completely accurate for a number of reasons: (1) there is no real world data to compare against, (2) there is little data, (3) real-world data is inaccurate, (4) even if the data is accurate, the real world data is only a sample, which in itself creates inaccuracy. Verification and Validation are continuous and iterative processes performed throughout the life cycle of a simulation study [70].

12. **Experimental design.** The experimental design improves the efficiency of the experimentation process. In this stage, the experimental factors that are most likely to lead to significant improvements are identified. This process is developed using data analysis, expert knowledge, preliminary experimentation and sensitivity analysis.

13. **Experimentation.** Experimentation is conducted following the experimental design guidelines. It can make use of multiple simulation replications; single long run (equivalent to taking one large sample in statistics); interactive experimentation (observing the simulation and making changes to the model to see the effects); batch experimentation (setting experimental factors and leaving the models to run for a pre-defined run length); comparing alternatives (where there is a limited number of scenarios to be compared) and search experimentation (when there is no predefined number of scenarios).

14. **Result Analysis.** Plots and statistics are collected during the simulation. The result analysis is the process that interprets results and the best way to present them.

15. **Report Findings.** After results are interpreted, there is the need to report the findings from the simulations.For immunology it can be new insights, verification of a theory, etc.

16. **Validate and add more requisites with immunologists.** Building an immune simulation is an iterative process. Generally the model is built together with immunologists, and, in every step of the framework, the model elements should be verified with them.

The process of simulation is iterative, as shown in Figure 3.1. During the model development, additional data might become available, which changes the system description/objectives and impacts on every step of the process. Moreover, as validation occurs throughout the whole process, if any of the stages is not validated (data available, real world understanding and description, conceptual model, computer model, experimental design, etc), there is the need to go back and rethink the invalid state, which impacts on the subsequent steps.

For our case studies, however, we will not be concerned with many of the steps outlined, as we are developing simulations from existing mathematical models. Instead, our focus will be on (1) the development of conceptual models for the SD and ABMS approaches; (2) on the representation of elements, parameters, etc. for both SD and ABMS; (3) on the building of the models; and (4) on the validation of the models and a comparison of outcomes.

A detailed explanation of how we developed our conceptual models, conducted our our experiments and validated our models will be introduced in the sections that follow.

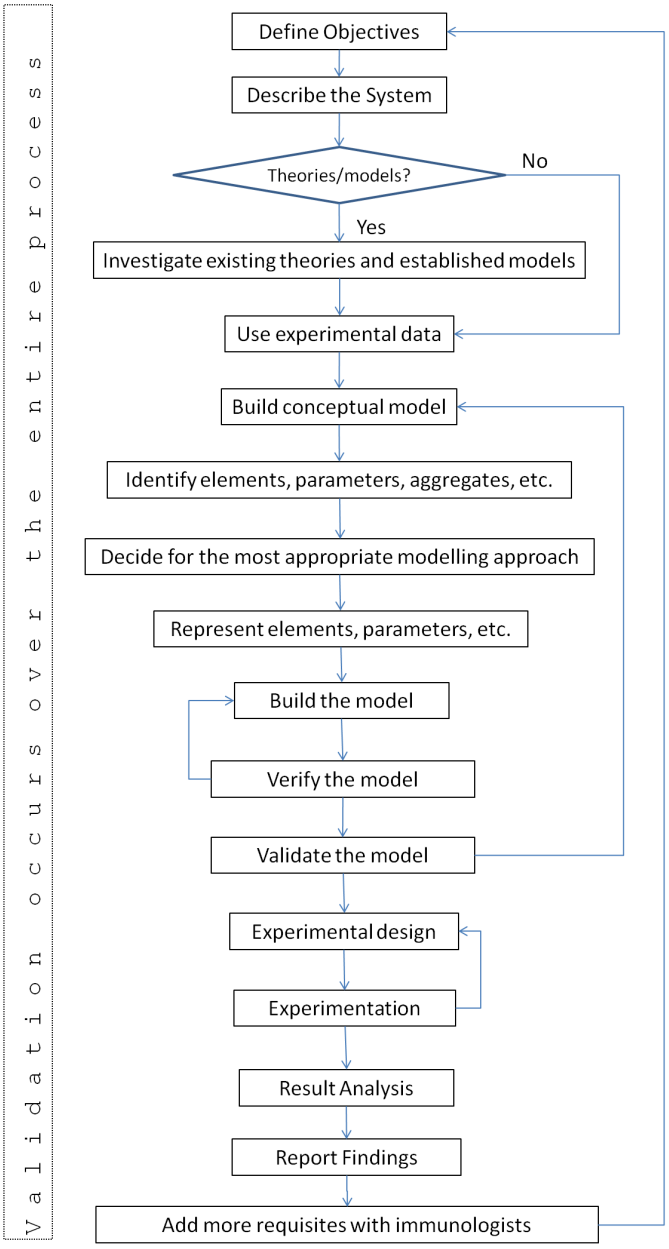


Figure 3.1: Process of simulation study: steps and their iterations.

3.4.3 Developing our Conceptual Models

Based on our case studies, SD and ABMS conceptual models are developed, representing the scope and level of the systems under investigation. The concept for the SD models is described in a stock and flow diagram, where we consider the causal-loop diagrams,

constant values and parameters for each simulation.

In addition, an individual-centric approach is used to represent our agents and their interactions. This approach is developed by using state charts and tables containing each agent description. The state charts show the possible different states of an entity and define the events that cause a transition from one state to another. As ABMS is an object-oriented approach and agents are object instantiations, each table includes the attributes, reactive and proactive behaviours of the objects.

3.4.4 Experimentation and Validation

As our case studies derive from mathematical models, we validated our SD models by comparing their outputs to those outputs produced by the mathematical models. We then validated our ABMS models by comparing their outputs to those outputs produced by the SD models (i.e. our base model for the comparison). We ran the simulations on an *Intel Core*^{TM} Duo CPU 2GHz and 2GB RAM.

As ABMS is a stochastic simulation method, we conducted several replications. We ran fifty replications for each case study and calculated the mean values for the outputs.

The samples obtained by SD and ABMS were statistically compared using the Wilcoxon rank-sum test to formally establish whether they are statistically different from each other. This test is applied, as it is robust when the populations are not normally distributed, which is the case of the samples obtained by the SD and ABMS. Other approaches for assessing whether the two samples are statistically different, such as the t-test, could provide inaccurate results as they perform poorly when the samples are non-normal. In addition to the statistical test, for some cases we also reported time and computational resources demanded in the simulations for each approach.

Our work also made use of case studies in order to validate the conversion guidelines introduced in the next section (Section 3.5). The experiments and case studies presented in this thesis were implemented using *AnyLogic*^{TM} 6.5 University version (XJTechnologies 2010) [95] and our conversion methodology was therefore tested using this tool.

3.5 Conversion between Approaches

As defined in Section 3.3, one objective of this thesis is to create and test translation guidance between approaches. Once an immune simulation is developed under a certain paradigm (following the steps defined in Section 3.4.2) we want to investigate if it is possible to obtain an equivalent model in another paradigm. In this section we introduce our own set of guidelines for this conversion, which will be further tested in the next chapters.

We start by showing the conversion from ODEs to SD (Section 3.5.1). Subsequently, we suggest guidance to convert from SD to ABMS (Section 3.5.2), from ODEs to ABMS(Section 3.5.3) and from ABMS to SD (Section 3.5.4). These guidelines were defined based on experiments we performed in [26–28]. For all the conversions, we also exemplify the conversion techniques applied to classical simulation problems found in the literature.

3.5.1 From Ordinary Differential Equations to System Dynamics

For our conversions to SD, we consider stock and flow diagrams instead of causal-loop diagrams, due to the fact that stock and flow diagrams provide information about different types of variables and their functions. We propose the following steps to perform this conversion, based on our case studies:

1. Identify the stocks. The stocks in the ODEs are the values that change/acumulate with time and which there is interest to keep information throughout the simulation. For instance, any X in $\frac{dX}{dt}$.

2. Identify stock's inflows. The inflows will be any calculation in the ODEs that increases the value of $\frac{dX}{dt}$.

3. Identify the stock's outflows. The outflows will be any calculation in the ODEs that decreases the value of $\frac{dX}{dt}$. For instance, if the ODE is $\frac{dX}{dt} = a - b$, a is an inflow whereas b is an outflow.

4. Determine the information. The information about the stocks is given by the use of these stock values in the flows calculations. The mathematics defining inflows and outflows are directly obtained from the ODEs, referred to in the previous step. For example, let us consider the ODE $\frac{dX}{dt} = aX - bY$ (X and Y are stocks, aX is the inflow, bY is the outflow, a and b are constant values defined in the mathematical equations for the flows). The equation demonstrates that there are causal relationships between the stock X and itself and between flows X and Y. This means that the value of X and Y with time will affect X. In the stock and flow diagram there should therefore be an information arrow from flow X to the inflow aX and another information arrow from flow Y to the outflow bY, as shown in Figure 3.2.

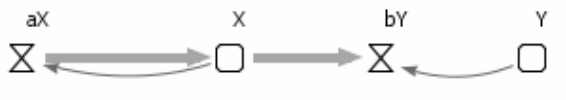


Figure 3.2: Example of information definition

When using a tool such as AnyLogic [95], once the inflows and outflows are expressed mathematically in the SD model, the information will appear automatically in the model stock and flow diagram.

5. Identify the parameters. The parameters will be any given value (constant or those that vary for each experiment). For example, in the equation $\frac{dX}{dt} = cX$, where $c = 2.3$, then c is a parameter in the SD.

Determine the flow calculations based on informations and parameters. In most cases, the value of a flow is based on the value of a stock and/or the value of one or more parameters. Hence, these variables have to be added in the flow formula. By looking at the mathematical equations and the information defined, it is possible to determine how the stocks and parameters should be placed in the formula. For other examples, however, a flow is determined just by mathematical expressions. Hence the correspondent expression from the mathematical model should be transferred to the flow calculation.

Example

In order to illustrate the conversion from a system of ODEs to SD, let us consider a classical example from ecology [85] that models the dynamics of two populations, one predator species y and one prey species x, described by the following equations:

$$\frac{dx}{dt} = (1 - y - \lambda x)x \tag{3.1}$$

$$\frac{dy}{dt} = \alpha(x - 1 - \mu y)y \tag{3.2}$$

In order to obtain an equivalent SD model, the guidelines defined above will be followed:

1. Identify the stocks: The stocks in the predator-prey example are x and y, as the ODEs describe their dynamics with time ($\frac{dx}{dt}$ and $\frac{dy}{dt}$).
2. Identify stock's inflows:
   - There is only one inflow for the stock x, which is $1 \times x$, as it adds to the stock variable.
   - Similarly, there is one inflow to the stock y, i.e. αxy.
3. Identify the stock's outflows:
   - The outflows for the x stock are xy and λx^2
   - For the stock y, the outflows are αy and $\alpha \mu y^2$
4. Determine the information:
   - All inflows and outflows use the value of the stock x in their calculations, therefore, there is information from this stock to all flows. Furthermore, the outflow xy uses information from the stocks x y, as illustrated in Figure 3.3.
   - Similarly to the stock x, there is information coming from Y to all its stocks, as shown in Figure 3.4.

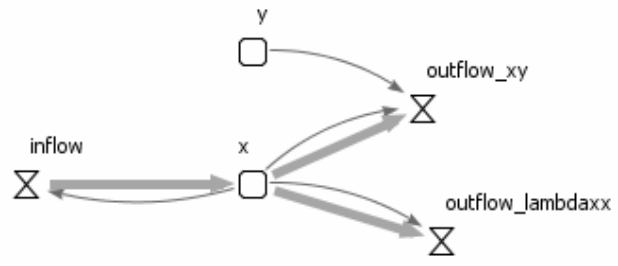


Figure 3.3: The stock x and its flows and information

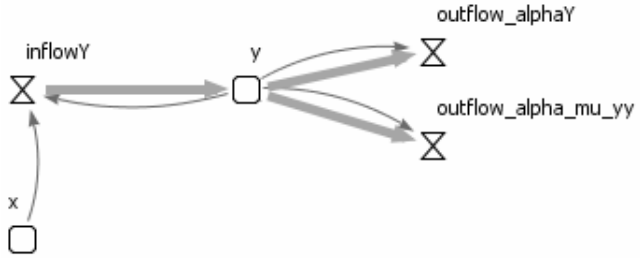


Figure 3.4: The stock y and its flows and information.

5. Identify the parameters: The parameters in the model are the constant values α, λ and μ.

6. Determine the flow calculations based on informations and parameters. By looking at Equations 3.1 and 3.2, it is possible to define the flow calculations, as depicted in Table 3.1. The final SD model is determined after the flow calculations are defined. The final stock and flow is shown in Figure 3.5.

Table 3.1: Flow calculations for the predator-prey example

Stock	Flow	Expression	Flow formula
x	$inflow$	x	x
	$outflow\_xy$	xy	xy
	$outflow\_lambdaXX$	λx^2	$lambdax^2$
y	$inflowY$	αxy	$alphaxy$
	$outflow\_alphaY$	αy	$alphay$
	$outflow\_alpha\_muyy$	$\alpha \mu y^2$	$alpha.mu.y^2$

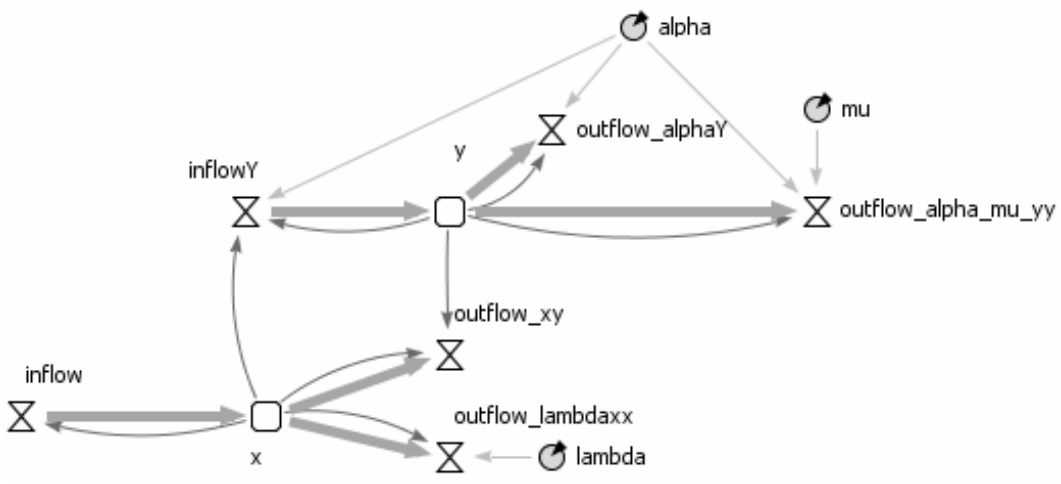


Figure 3.5: The final SD model for the predator-prey example

3.5.2 From System Dynamics to Agent-based Modelling and Simulation

In order to convert the SD into an ABMS model, we propose the following steps [27]:

1. Identify the possible agents. For this purpose, we use some characteristics defined in [53]. An agent is: (1) self-contained, modular, and a uniquely identifiable individual; (2) autonomous and self-directed; (3) a construct with states that varies over time; and (4) social, having dynamic interactions with other agents that impact its behaviour. By looking at the SD stock and flow diagram, therefore, the stocks (their disaggregation) will either be corresponding to agents or states of one agent [26]. The decision whether the stock is an agent or an agent state varies depending on the problem investigated. Based on our case studies presented on the next chapters, however, we suggest that:

- stocks preferably become states when they represent accumulations of elements from the same population.
- stocks become agents when they represent accumulations from different populations.

2. Identify the behaviour and rules of each agent. As we are building ABMS models from SD models, the agent's behaviours will be determined by mathematical equations converted into rules. Each agent has two different types of behaviours: reactive and proactive behaviours. The reactive behaviour occurs when the agents perceive the context in which they operate and react to it appropriately. The proactive behaviour describes the situations when the agent has the initiative to identify and solve an issue in the system.

3. Implement the agents. Based on the conceptual model derived from step 2 we develop state charts, one for each agent type. The state charts model states and state transitions. Moreover, at this stage, the behaviours of each agent are implemented using the simulation tool.

4. Build the simulation. After agents are defined, their environment and behaviour previously established should be incorporated in the simulation implementation. Moreover in this step we include parameters and events that control the agents or the overall simulation.

Example

In order to show a practical application of our guidelines, let us consider a classical SD model, namely, the bass diffusion model [83]. This is a model of a product diffusion, where there are two stocks representing potential adopters and adopters of the product. Potential adopters become adopters at a certain adoption rate. This rate depends on advertisement and word of mouth promotion. The ODEs for each stock are defined as below:

$$\frac{dPotentialAdopters}{dt} = -AdoptionRate \tag{3.3}$$

$$\frac{dAdopters}{dt} = AdoptionRate \tag{3.4}$$

where:

- $AdoptionRate = AdoptionFromAdvertising + AdoptionFromWordOfMouth$
- $AdoptionFromAdvertising = AdvertisingEffectiveness * PotentialAdopters$
- $AdoptionFromWordOfMouth = ContactRate * AdoptionFraction * \frac{PotentialAdopters * Adopters}{PotentialAdopters + Adopters}$

The stock and flow diagram for the bass diffusion model is therefore shown in Figure 3.6. In the model, we split the *AdoptionRate* in two inflows, *AdoptionFromAdvertising* and *AdoptionFromWordOfMouth*:

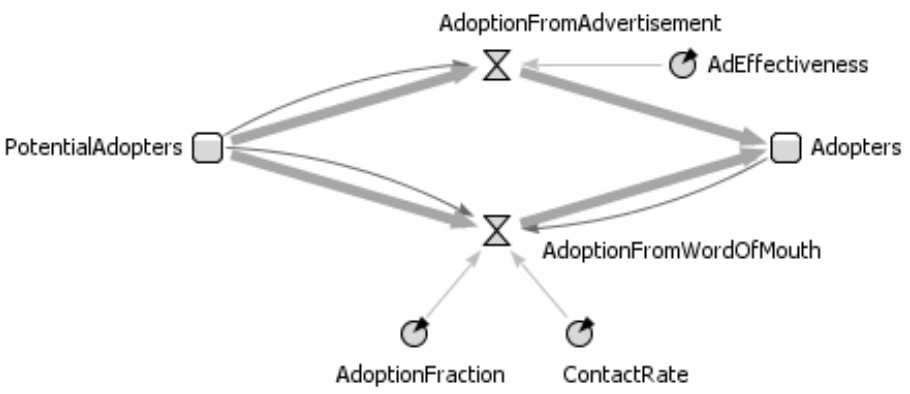


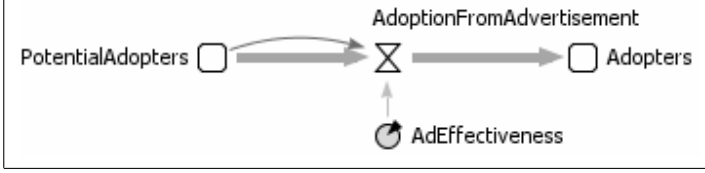
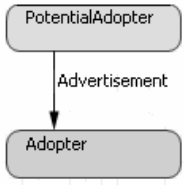
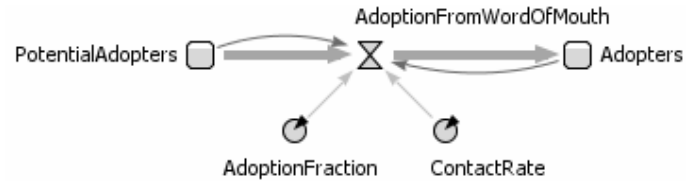
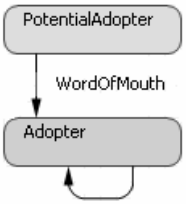
Figure 3.6: The SD model for bass diffusion

In order to obtain an equivalent ABMS model, we will use the guidelines defined in this section:

1. Identify the possible agents: by looking at the SD stock and flow diagram, as the stocks represent accumulations of elements from the same population, there will be only one agent, which can assume the *Adopters* or *PotentialAdopters* state.
2. Identify the behaviour and rules of each agent: as for this example there is only one agent that changes state at a point in time, the behaviour is to become an adopter according to a certain rate, if the current state is PotentialAdopter.

3. Implement the agents: the flows were then converted to the appropriate transition. In order to build the equivalent element in the state chart given the flow information, the correspondence diagram shown in Figures 2.8 and 2.9 on Section 2.5.2 of the previous chapter was used. The ABMS correspondents are therefore shown in Table 3.2. In the table, the first column contains the SD stock and flow diagrams for each flow *AdoptionFromAdvertisement* and *AdoptionFromWordOfMouth*. The second column presents the corresponding transitions from the *PotentialAdopter* state to the *Adopter* state. The *Advertisement* transition was obtained by using case B of Figure 2.8 (page 31); the transition $WordOfMouth$ was obtained using case C of Figure 2.9 (page 32).

Table 3.2: SD flows converted into ABMS

System Dynamics	Agent-based
	
	

The final stock and flow diagram for the adopting agent is shown in Figure 3.8.

4. Build the simulation: the final simulation model will contain a set of agents as described above that will turn from potential adopters to adopters, according to the rates pre-defined by the system user.

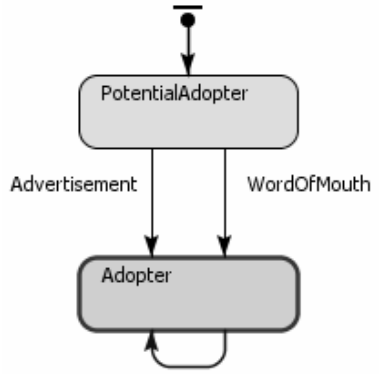


Figure 3.7: The bass diffusion agent

3.5.3 From Ordinary Differential Equations to Agent-based Modelling and Simulation

The process of conversion from ODEs to ABMS is quite similar to that showed for the conversion from SD to ABMS (Section 3.5.2), as shown in the next steps:

1. Identify the possible agents. By looking at the ODE equations, the variables differentiated in time (for example, X in $\frac{dX}{dt}$) will either be corresponding to agents or states of one agent. The decision whether the variable is an agent or an agent state varies depending on the problem investigated. Based on our case studies presented on the next chapters, however, we suggest that:

- the variables differentiated in time preferably become states when they represent elements from the same population.
- variables differentiated in time become agents when they represent different populations.

2. Identify the behaviour and rules of each agent. As we are building ABMS models from ODE models, the agent's behaviours will be determined by mathematical equations converted into rules.

3. Implement the agents. Based on the conceptual model derived from step 2 we develop state charts, one for each agent type. The state charts model states and state transitions. Moreover, at this stage, the behaviours of each agent are implemented using the simulation tool.

4. Build the simulation. After agents are defined, their environment and behaviour previously established should be incorporated in the simulation implementation. Moreover in this step we include parameters and events that control the agents or the overall simulation. The value for these parameters is also obtained from the ODEs.

Example

Let us consider once again the ODE-based bass diffusion model [83]:

$$\frac{dPotentialAdopters}{dt} = -AdoptionRate \tag{3.5}$$

$$\frac{dAdopters}{dt} = AdoptionRate \tag{3.6}$$

where:

- $AdoptionRate = AdoptionFromAdvertising + AdoptionFromWordOfMouth$
- $AdoptionFromAdvertising = AdvertisingEffectiveness * PotentialAdopters$
- $AdoptionFromWordOfMouth = ContactRate * AdoptionFraction * \frac{PotentialAdopters * Adopters}{PotentialAdopters + Adopters}$

In order to obtain an equivalent ABMS model, we will use the guidelines defined in this section:

1. Identify the possible agents: by looking at the ODEs, there will be only one agent, which can assume the *Adopters* or *PotentialAdopters* state.

2. Identify the behaviour and rules of each agent: the identified behaviour is to become an adopter according to a certain rate, if the current state is Potential-Adopter.

3. Implement the agents: The process to implement the agents is similar to that shown in the previous section. The main difference is that instead of considering the flows, the mathematical equations should be used. The final stock and flow diagram for the adopting agent is shown in Figure 3.8.

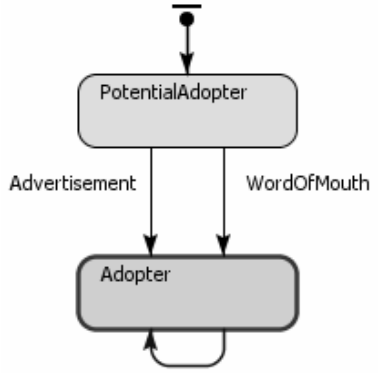


Figure 3.8: The bass diffusion agent

4. Build the simulation: the final simulation model will contain a set of agents as described above that will turn from potential adopters to adopters, according to the rates pre-defined in the ODE model.

Although we showed the steps to convert from ODEs to ABMS, these guidelines will not be further explored in our work, as our main focus of interest lies in the systems simulation methods SD and ABMS.

3.5.4 From Agent-based Modelling and Simulation to System Dynamics

For converting the ABMS model into an SD model, we propose the following steps [27]:

1. Identify the system structure. First we have to recognize the system structure and assume a high level of aggregation for the objects being modelled. It is necessary to generalise from the specific events and consider patterns of behaviour that characterise the situation. The system autonomous elements will therefore no longer respond individually. The simulation outcome will be given by the collection of individuals and its dynamics as a group.

2. Identify the stocks in the system. Stocks are physical entities which can accumulate over time.

3. Define the stocks and their flows. Having the stocks (step 2) and the information about the structure of the model (step 1) we can depict how each stock is changed over time by the flows and the information about how a stock would influence a flow.

4. Define the final stock and flow diagram. After defining the diagrams for each stock, it is necessary to go back to the system structure and define how the stocks will interact or influence each other.

5. Define the mathematical model. For SD, a set of mathematical equations is necessary to describe how the stocks will change over time. In our case studies, the information provided by the ABMS is not enough to build an SD model because we do not have the equations and rates defining the dynamics of each population. Therefore, to continue building the model we need extra information. For example, a data set or a well-established model that describes mathematically how the system changes over time would be necessary. In addition, there are also cases where further parameter calibration is necessary.

6. Define the parameters of the mathematical model. In our case studies, the parameters were obtained from the state charts transition rates.

7. Define the flow calculations. In our case studies, a flow expression is defined by looking at the transition rate calculation and the information defined in the stock and flow diagram.

8. Define the final SD model. The final SD model is supposed to contain the complete stock and flow diagram together with the flow calculations and parameters used in the calculations.

Example

In order to show the conversion from ABMS to SD, let us consider a classical viral spread model, namely SIR, where a person can assume three states: susceptible, infectious or recovered. When in the infectious state, a person can infect another person, as shown in the state chart of Figure 3.9:

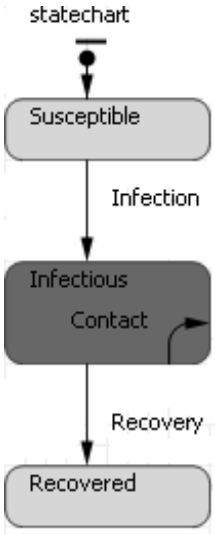


Figure 3.9: The SIR agent

The simulation consists of a set of agents and, initially, only one agent is in the state infectious whereas the remaining agents are in the state susceptible. The transition infection is triggered by a message from another agent in the state infectious, and the message will be sent according to a rate in which the transition contact is triggered. In the model, this rate is defined by the formula $\frac{ContactRate}{InfectionProbability}$, where the values of $ContactRate$ and $InfectionProbability$ are constant. A person's recovery is determined by the rate $\frac{1}{AverageIllnessDuration}$, where the value $AverageIllnessDuration$ is also constant. This rate triggers the transition recovery in the state chart.

In order to obtain an equivalent SD model, we will follow the steps proposed:

1. Identify the system structure: for the SD, as we look at aggregates instead of individuals, there are three populations to be considered, i.e. the individuals susceptible to the infection, the population of infected people and those individuals who recovered from the infection.

2. Identify the stocks, flows, and information in the system: the stocks will be the susceptible, the infectious and the recovered populations. There is an outflow from the stock susceptible to the stock infectious and another outflow from the stock infectious to the stock recovered, as shown in Figure 3.10.

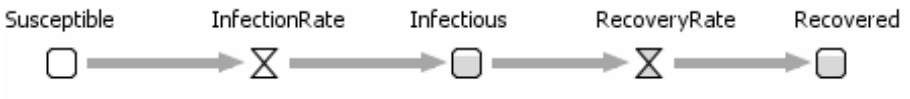


Figure 3.10: The SD model stocks and flows for the SIR model

The susceptible population decreases according to to the infections occurred. So there is the need of information about both susceptible and infectious stocks in the flow $InfectionRate$. As only the infectious population has influence in the population recovery, there is information from the stock infectious to the flow $RecoveryRate$, as shown in Figure 3.11. As this is a simple model, this figure illustrates the final stock and flow diagram.

Figure 3.11: The SD model stocks and flows and information for the SIR model

3. Define the mathematical model: the final model is described by the following equations:

$$\frac{dSusceptible}{dt} = Susceptible * Infectious * \frac{ContactRate}{(TotalPopulation) * InfectionProbability} \tag{3.7}$$

where the term $\frac{ContactRate}{InfectionProbability}$ was obtained from the transition *Contact* in the state chart, observing the conversion guides of Figures 2.8 and 2.9. As this transition occurs for each agent in the *Infectious* state, when converting to SD, there is the need to multiply the value in the flow by the population of infectious, which is the *Infectious* stock. As now all population is considered, the probability infection needs to be distributed throughout the sum of the three populations. Hence, the variable $TotalPopulation$ is included in the calculation. It is not trivial to spot the need of the variable $TotalPopulation$. However, a final model calibration to obtain similar results as those from the ABMS would produce this value.

$$\frac{dInfectious}{dt} = InfectionRate - RecoveryRate \tag{3.8}$$

$$\frac{dRecovered}{dt} = \frac{Infectious}{AverageIllnessDuration} \tag{3.9}$$

where the rate $\frac{1}{AverageIllnessDuration}$ was obtained from the *Recovery* transition rate calculation.

4. Define the parameters of the mathematical model: the parameters extracted from the mathematical equations are $ContactRate$, $InfectionProbability$ and $AverageIllnessDuration$, which values are the same as those from the ABMS model and should be used in the flow calculations. The final SD model, with the stock and flow diagram and parameters is shown in Figure 3.12:

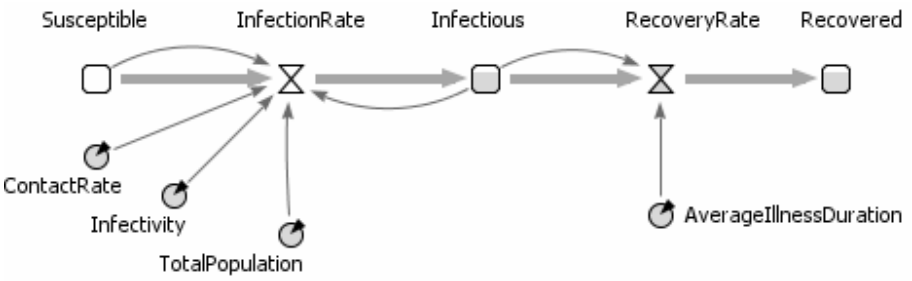


Figure 3.12: The final SD for the SIR model

3.6 Summary

This chapter describes the research methodology defined in order to achieve the research objectives of this thesis. In choosing the case study methodology to conduct investigations, the baseline of our models consists of established ODE models that describe some immune mechanisms. We consider aspects such as the behaviour of the entities of the model, the type of hypothesis to be tested, the empirical embeddedness of real data and the modelling effort for the comparison of the approaches. For each aspect, we use a multiple-case approach comprising three case studies, which will be further introduced in subsequent chapters. Furthermore, in this chapter we introduce our own set of guidelines for the conversion from ODEs to SD, SD to ABMS, ODEs to ABMS and ABMS to SD. These guidelines will be used and tested on our case studies, which are presented in the following chapters.

Chapter 4

Static Non-spatial models

4.1 Introduction

In this chapter simulations in immunology are developed as part of the fulfilment of the objectives introduced in Section 3.3. In particular, the goals targeted are:

1. To test the guidelines defined to convert from ODE to SD and to convert from SD and ABMS models and assess the impact of this conversion on models involving static non-spatial entities.

2. To compare SD and ABMS outcomes considering static non-spatial entities in the model, the type of hypothesis to be tested, the empirical embeddedness of real data and the modelling effort.

3. To define guidance to choose between SD and ABMS depending on the characteristics of the problem to be addressed.

Simulations are built taking into account three different case studies based on mathematical modelling, in which we test the conversion guidelines, compare SD and ABMS outcomes and assess the impact of the conversions. These case studies were chosen based on characteristics such as population size, modelling effort, model complexity, observation of the ODEs outcome results and the number of different populations modelled.

The mathematical models chosen vary largely within these aspects and therefore we can perform a more robust analysis on the effectiveness of our guidelines.

The first case study – which is the most complex model in terms of population sizes and elements considered – is based on an ODE model involving interactions that influence naive T cell populations with age (Section 4.2). For the second case study, which is the simplest one with only one population, we investigate mathematical models of general tumour growth (Section 4.3). Our third case study – which also complex in terms of different populations – comprises an ODE model of cell-free viral spread of the human immunodeficiency virus (HIV) in the bloodstream (Section 4.4). In Section 4.5, we discuss the results obtained and draw the conclusions for our first set of experiments.

4.2 Case 1: Naive T Cells Output

In this section we investigate our first case study, which compares SD and ABMS for an immune system ageing model that involves interactions which influence the naive T cell populations over time. The model is based on the mathematical equations defined in [59]. In their work, Murray *et al.* [59] propose a model with a set of equations to fit observed data and estimate the output of a certain type of immune cells with age. These cells are the naive T cells and play an important role in the immune system by responding to new infections. With age, these responses become less frequent and ineffective. This age-associated problem occurs because the organism lacks naive cells derived from the thymus, as a result of thymic shrinkage over time. As a consequence, the naive repertoire changes from the thymic source to the peripheral proliferation source. Thus, there is no new phenotypical naive cell entering the system. Further details on thymus output shrinkage and its impact on naive T cells population will be given in sections 4.2.1 and 4.2.2.

The remainder of this section is organised as follows. In Section 4.2.3 the mathematical model regarding the naive T cells population dynamics is introduced. Section 4.2.4

shows how to obtain an equivalent SD model from the ODEs. In Section 4.2.5, we apply our conversion guidelines to obtain an ABMS model from the SD. The experimental design is defined in Section 4.2.6 and the results are presented in Section 4.2.7. Finally, a summary of the case study and findings is given in Section 4.2.8.

4.2.1 Lack of naive T cells

Before an individual reaches the age of 20, the set of naive T cells is sustained primarily from thymic output [59]. In middle age, however, there is a change in the source of naive T cells: as the thymus involutes, there is a considerable shrinkage in its T cell output, which means that new T cells are mainly produced by peripheral expansion. There is also a belief that some memory cells have their phenotype reverted back to the naive cells type [59].

These two new methods of naive T cell repertoire maintenance, however, are ineffective [59] as they do not produce new phenotypic changes in the T cells. Rather, evidence shows that they continue to fill the naive T cell space with copies of existing cells [55]. The loss of clones of some antigen-specific T cells therefore becomes irreversible. These age-related phenomena lead to a decay of immune performance in fighting aggressors.

4.2.2 Naive T cell output

Thymic contributions in an individual are quantified by the level of a biological marker known as 'T cell receptors excision circle' (TREC). TREC is circular DNA originated during the formation of the T-cell receptor. The percentage of T cells possessing TRECs decays with shrinkage of thymic output and activation and reproduction of naive T cells [59]. This means that naive T cells originating from the thymus have a greater percentage of TREC than those originating through other proliferation.

The first case study is based on data and equations obtained in [59], which are concerned with establishing an understanding of naive T cell repertoire dynamics. The objective of the model is to determine the likely contribution of each of the naive T cell's sources

by comparing estimates of the presence of TREC in the cells (see Figure 4.1 below). The dynamics of the sustaining sources, i.e. naive proliferation, TREC and reversal of memory to naive T cells, are modelled mathematically.

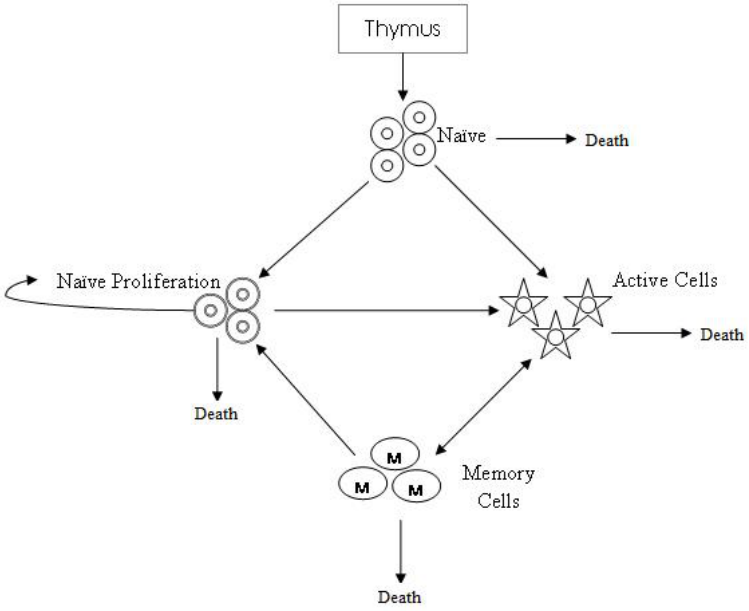


Figure 4.1: Dynamics of Naive T cells

4.2.3 The Mathematical Model

The mathematical model proposed in [59] is described by equations 4.1 to 4.6, in which N is the total number of naive cells of direct thymic origin, N_p is the number of naive cells that have undergone proliferation, A is the number of activated cells, M is the number of memory cells and t is time (in years). At the beginning of life the main source of naive T cells is the thymus and therefore most naive T cells in the body belong to the population N. With time naive T cells from thymus proliferate, which contributes for the increase of the N_p population. When the immune system faces a new threat, naive T cells are recruited and become active (A). A fraction of active cells turns into memory cells (M).

The first differential equation describing the naive T cell population from thymus is:

$$\frac{dN}{dt} = s_0 e^{-\lambda_t t} s(N_p) - [\lambda_n + \mu_n g(N_p)]N \tag{4.1}$$

where:

- s_0 is the thymic output value
- λ_t is the thymic decay rate
- t represents time in years
- $s_0 e^{-\lambda_t t} s(N_p)$ represents the number of cells that arise from the thymus
- $s(N_p)$ is the rate of export of the thymus defined by:

$$s(N_p) = \frac{1}{1 + \frac{\bar{s}^{N_p}}{\bar{N}_p}} \tag{4.2}$$

- $\bar{N}_p$ and $\bar{s}$ are equilibrium and scaling values respectively. These values were defined in the experiments in [59].
- $\lambda_n N$ represents the naive cells that become part of the naive proliferating population
- λ_n is the naive proliferation rate
- μ_n is the thymic naive cells death rate
- $\mu_n g(N_p)N$ represents the naive cell death rate
- $g(N_p)$ is the death rate between naive TREC-positive and naive TREC-negative cells, defined as:

$$g(N_p) = 1 + \frac{\frac{bN_p}{\bar{N}_p}}{1 + \frac{N_p}{\bar{N}_p}} \tag{4.3}$$

The second differential equation describing the naive T cells from proliferation is:

$$\frac{dN_p}{dt} = \lambda_n N + [ch(N, N_p) - \mu_n]N_p + \lambda_{mn} M \tag{4.4}$$

where:

- c is the proliferation rate
- $ch(N, N_p)N_p$ represents the naive proliferation
- $h(N, N_p)$ is the dilution of thymic-naive through proliferation defined by:

$$h(N, N_p) = \frac{1}{1 + \frac{N+N_p}{\tilde{N}_p}} \tag{4.5}$$

- $\mu_n N_p$ is the death rate of proliferation-originated naive cells
- λ_{mn} is the reversion rate from memory into N_p

The final differential equation for the memory cell population dynamics is:

$$\frac{dM}{dt} = \lambda_a A - \mu_m M - \lambda_{mn} M \tag{4.6}$$

where:

- λ_a is the reversion rate into memory
- μ_m is the death rate of memory cells

The parameter values for the model can be seen in Table 4.1.

For the mathematical model and subsequent simulations, $s0 = 56615$. The values for active cells over time are determined by referring to data collected by [14] (Figure 4.2 below). This table contains the number of activated CD4+ cells per mm^3 for early years

Table 4.1: Rate values for the mathematical model (obtained from [59])

rate	value(s)
λ_t	$\frac{log(2)}{15.7}(year^{-1})$
λ_n	0.22, 2.1, 0.003
μ_n	4.4
c	0 (no proliferation) or $\mu_n(1+\frac{300}{N_p})$
λ_{mn}	0
μ_m	0.05
λ_a	1

and is used as a stock for the active cells. From the active cell stock the values of the memory cell stock are updated according to the parameter λ_a.

Equations 4.1 to 4.6 are incorporated in the SD and ABS models in order to investigate if it is possible to reproduce and validate the results obtained in [59]. Moreover, variations of the ratio variables are explored to understand the importance of each individual integrand in the system. For example, it is important to establish how much the proliferation rate impacts on the depletion of naive T cells over age, and to identify the point in time at which the system can be defined as losing functionality.

4.2.4 The System Dynamics Model

From Ordinary Differential Equations to System Dynamics

The system dynamics model objective is to simulate the processes involved in the maintenance of naive T cells. The model is built taking into consideration all the interactions described by the mathematical equations defined in the previous section. Hence, we use the conversion from ODEs to SD guidelines introduced in Section 3.5.1:

1. Identify the stocks
2. Identify stock's inflows and outflows
3. Determine the information
4. Identify the parameters

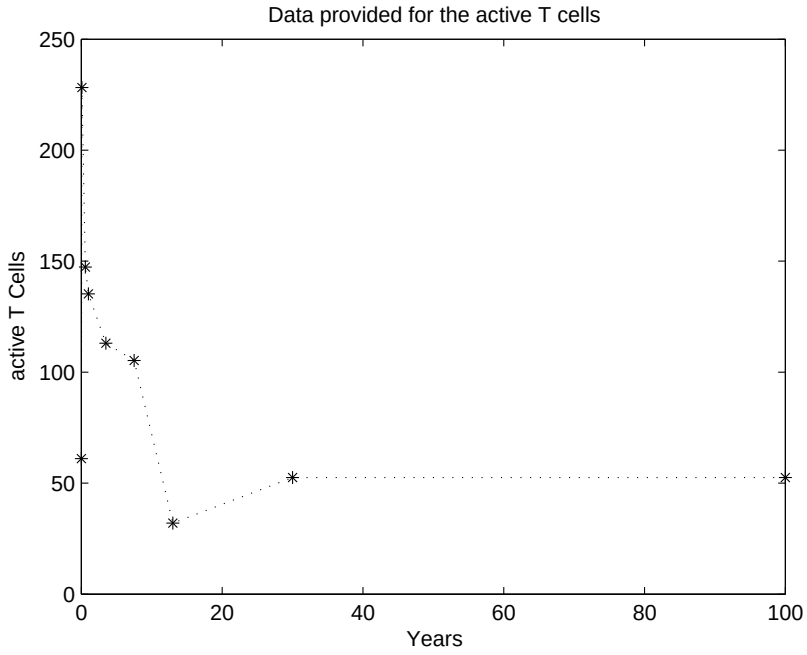


Figure 4.2: The data set used as a look-up table for the active cells. The data set contains the number of activated $CD4$ cells per mm^3 for early years taken from Comans-Bitter *et al.* [14].

5. Determine the flow calculations based on information and parameters

Model Stocks

The naive T cells, naive T cells from proliferation and memory cells are stock variables, as the aim is to keep information of how they accumulate over time.

Model Flows

The stock variable that represents the number of naive T cells is subject to inflowing thymic output ($ThymusOutput$), and proliferation ($NaiveCellsInProliferation$) and death ($NaiveDeath$) outflows. The flows between naive cells and active cells are not defined in the mathematical model. It is assumed, therefore, that these flows only interfere on the stock of active cells, which are not considered in the SD model. The

number of active cells, which is a stock, is given in a look-up table containing real values of active cells in the human organism. The graphical representation (stock and flow diagram) of the stock of naive T cells and its flows, corresponding to Equation 4.1 on the mathematical model, can be seen in Figure 4.3:

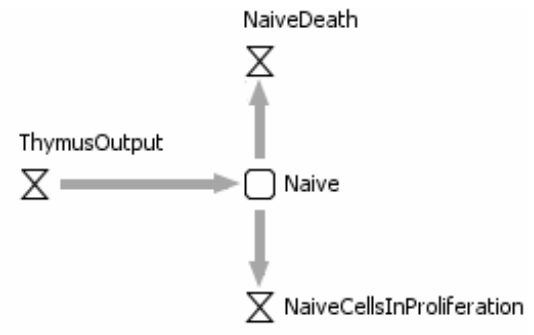


Figure 4.3: The naive T cell stock variable and its flows: thymic output, proliferation and death

The stock of naive cells from proliferation's inflows are proliferation and reversion from memory, and the outflow is death, according to Equation 4.2. The stock and flow diagram is shown in Figure 4.4.

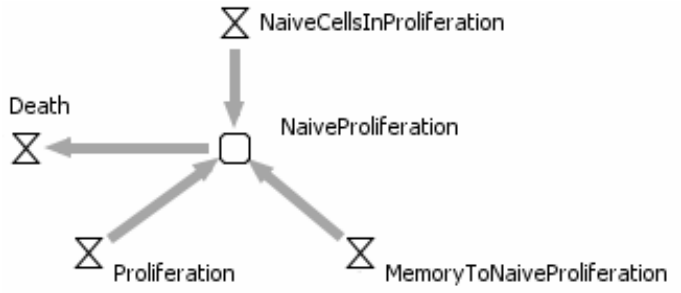


Figure 4.4: The naive T cells from proliferation stock variable and its flows: death, activation, proliferation and memory reverted to naive

The memory stock's inflow is reversion from active to memory cells (in Figure 4.5, $ReversionToMemory$). The outflows are reversion to a naive phenotype (in the figure, *MemoryToNaiveProliferation* and death, as defined by Equation 4.6).

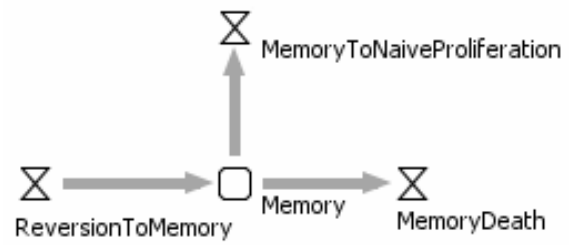


Figure 4.5: The memory T cells stock variable and its flows: reversion to a naive phenotype, reversion from active and death

Model Information

The next step for the conversion is to identify the information between the stocks. As we mentioned in Section, the SD's stock and flow diagram is constituted by stocks, flows, information, auxiliaries and parameters. Information (curved arrows in the stock and flow diagram) between stocks and flows indicates that there is an information about a stock that influences a flow (for further information refer to page 17).

By looking at Equation 4.1, it is possible to determine that there is information between the stock $Naive$ (N in Equation 4.1) and the flow $NaiveDeath$ and between the stock $Naive$ and the stock $NaiveProliferation$ (Np). For our implementation, functions are designed for s and g, which use the stock variable $NaiveProliferation$ in their calculations. Hence, the information about $NaiveProliferation$ is implicit in these functions. For the stock and flow diagram, therefore, there is information, which is from stock $Naive$ to the flow $NaiveDeath$, as show in Figure 4.6 below.

For the $NaiveProliferation$ stock there is information from it to $Proliferation$ and $Death$ flows, as shown in Figure 4.7.

In the $Memory$ stock there is information from it to the flows *MemoryToNaiveProliferation* and $MemoryDeath$, as shown in Figure 4.8.

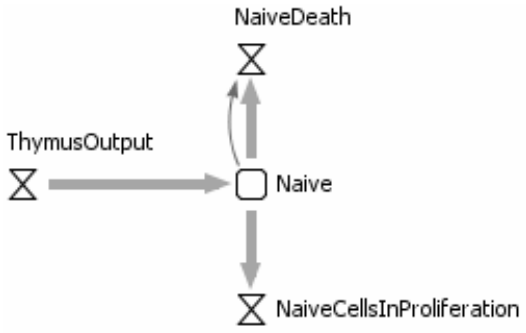


Figure 4.6: The naive T cell stock variable with its flows and information

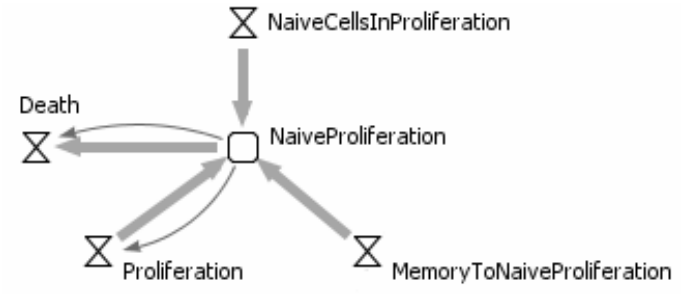


Figure 4.7: The naive T cells from proliferation stock variable with its flows and information

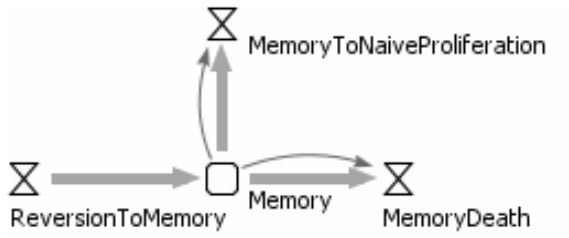


Figure 4.8: The memory T cells stock variable with its flows and information

Model Parameters

The model parameters are the same as those in the mathematical model. The mathematical parameters and their correspondents in the SD model are shown in Table 4.2:

Table 4.2: Parameters from the mathematical model and their correspondents from the SD model

rate	correspondent
s_0	$s0$
λ_t	$lambda\_t$
c	c
$\overline{N}_p$	bar_Np
$\overline{s}$	bar_s
b	n
λ_n	$ProliferationRate$
μ_n	$NaiveDeathRate$
λ_{mn}	$MemoryToNPRate$
μ_{N_p}	$NPDeathRate$
μ_m	$MemoryDeathRate$
λ_a	$ReversionToMemoryRate$

Flows Calculations

Table 4.3 demonstrates the flows for each stock, their correspondent in the mathematical model and the flow formula. In the table, the functions $s0()$, $s()$, $g()$ and $h()$ are implemented according to corresponding mathematical functions. The function $time()$ returns the current simulation time, which, for this case, is given in years. Furthermore, the $ThymusOuput$ is an example of flow which does not have any information or parameter. Hence, it is defined according to the mathematical expression stated.

The Final System Dynamics Model

All the stocks put together with flows, parameters and functions defined in the mathematical model form the SD model shown in Figure 4.9.

Table 4.3: Flow calculations for the naive T cell output model

Stock	Flow	Expression	Flow formula
Naive	*ThymusOutput*	$s_0 e^{-\lambda_t t} s(N_p)$	$s0()e^{-\lambda_t . time()} s()$
	NaiveCellsInProliferation	$\lambda_n N$	*ProliferationRate.Naive*
	NaiveDeath	$\mu_n g(N_p) N$	(*NaiveDeathRate.* $g()Naive$)
NaiveProliferation	*Proliferation*	$ch(N, N_p)$	($c \times h()$. *NaiveProliferation*)
	Death	$\mu_n N_p$	(*NpDeathRate.* *NaiveProliferation*)
Memory	*MemoryToNaiveProlife–ration*	$\lambda_{mn} M$	(*MemoryToNPRate.* *Memory*)
	ReversionToMemory	$\lambda_a A$	(*ReversionToMemoryRate.* *RealActives*(*time*()))
	MemoryDeath	$\mu_m M$	(*MemoryDeathRate.* *Memory*)

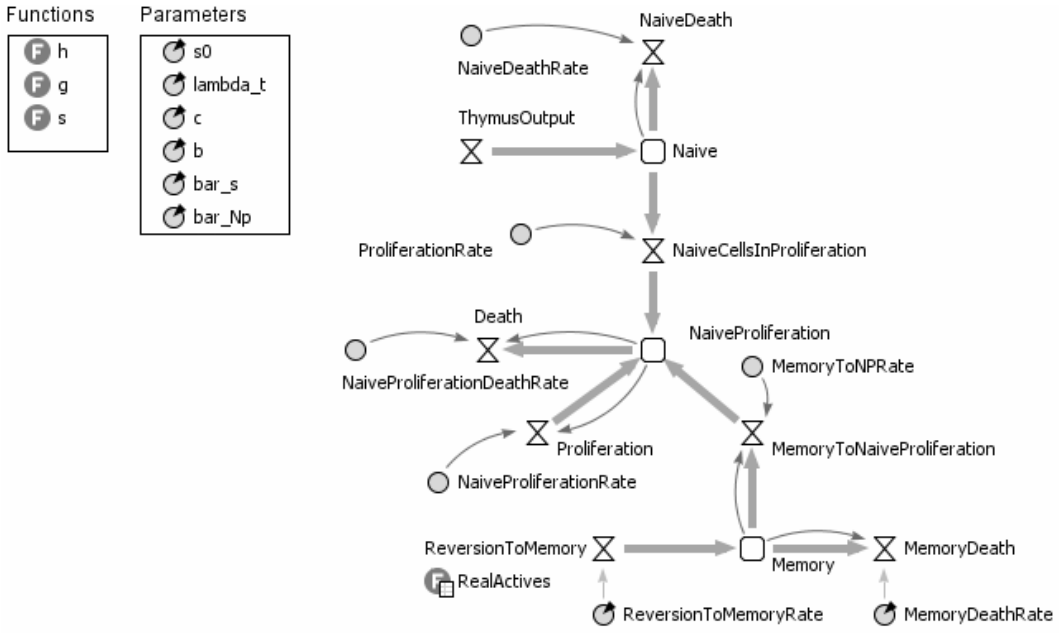


Figure 4.9: The system dynamics model's functions and parameters for case 1. The table function RealActives returns the values for active CD4+ cells at a certain time.

4.2.5 From System Dynamics to Agent-based Modelling and Simulation

In order to convert from SD to ABMS, the guidelines introduced in Section 3.5.2 are used:

1. Identify possible agents
2. Identify the behaviour of each agent
3. Implement the agents
4. Build the simulation

Model Agent

For the definition of the agents, by looking at the SD stock and flow diagram (Figure 4.2.4) there are two possible agent implementations:

1. Consider each stock variable ($Naive$, $NaiveProliferation$, $Memory$) as an agent or
2. Consider each stock variable as a different state of the same agent T cell

For the implementation, the second option was selected, i.e. T cells are the agents of the model. The reason each stock has not been implemented as a different agent is that it would be the same object duplicated. The main characteristics of the stocks are the same and their flows represent the same patterns of change in the accumulations (proliferation, death and source). A T cell agent, therefore, can assume three different states: $Naive$, $NaiveFromProliferation$ and $Memory$, as shown in the state chart depicted in Figure 4.10. The lozenge in the state chart represents a branch for the decision of the T cell current state:

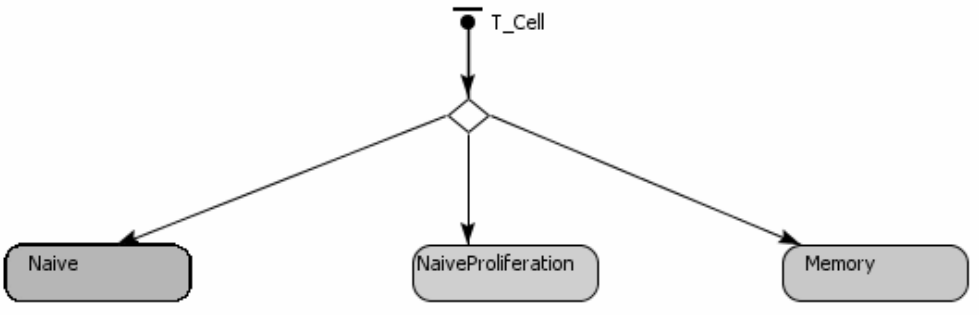


Figure 4.10: T cell agent states

Agent Behaviours

Each agent behaviour is driven by its current state and occurs according to a certain parameter rate. The agent's parameters and behaviours corresponding to each state are shown in Table 4.4 below. In this example, all behaviours are derived from the flows in the SD stock and flow diagram.

Table 4.4: Agents' parameters and behaviours for the naive T cell output model

State	Parameters	Reactive behaviour	Proactive behaviour
Naive	$NaiveDeathRate$	Dies	
		Is produced by thymus	
	$ProliferationRate$		Reproduces
NaiveProliferation	$NaiveProliferationDeathRate$	Dies	
	$ProliferationRate$	Is produced by Naive proliferation	
	$MemoryToNPRate$	Is produced from Memory	
	$NaiveProliferationRate$		Reproduces
Memory	$MemoryDeathRate$	Dies	
	$ReversionToMemoryRate$	Is produced from active cells	
	$MemoryToNPRate$		Turns into Naive

Although the thymic output determines the rate in which naive T cells are produced, this rate changes over time, and is therefore not considered as a parameter in our implementation. Instead, it is a dynamic variable.

Agent Implementation

The agent final state chart is depicted in Figure 4.11 below. T cells are the agents and the state chart represents all the states in which these agents can exist, i.e. naive, naive from proliferation or memory cells. There is also the final state when cells die and are eliminated from the system. The agents' state changes and their death rates are given by the ratios defined in the mathematical model. For instance, $NaiveDeathRate$ is equal to $\mu_n \times g()$ (Equation 4.1). Initially, all the agents are in the *naive* state. As the simulation proceeds, they can assume other stages according to the transition pathways defined in the state chart.

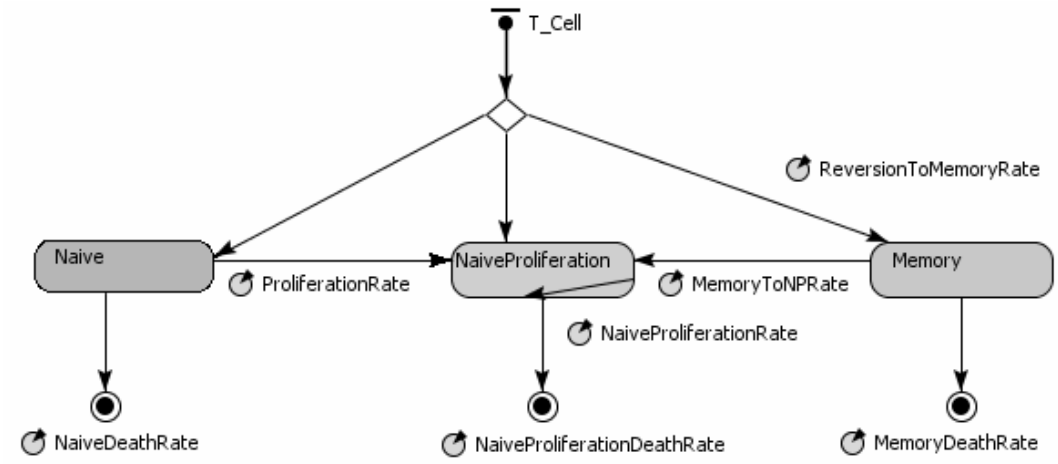


Figure 4.11: The T cell agent

When agents reproduce, the newborn agents, which are also T cells, should assume the same state as their original agent. Apart from proliferation, new agents are also produced from thymic output and reversion from active to memory cells. The algorithm that determines the agent state is given according to the flow chart in Figure 4.12. Its definition is based on the stock and flow diagram inflows for each stock.

The agents die according to specific rates determined by the mathematical model. The agents in this simulation respond to changes in time and do not interact with each other directly.

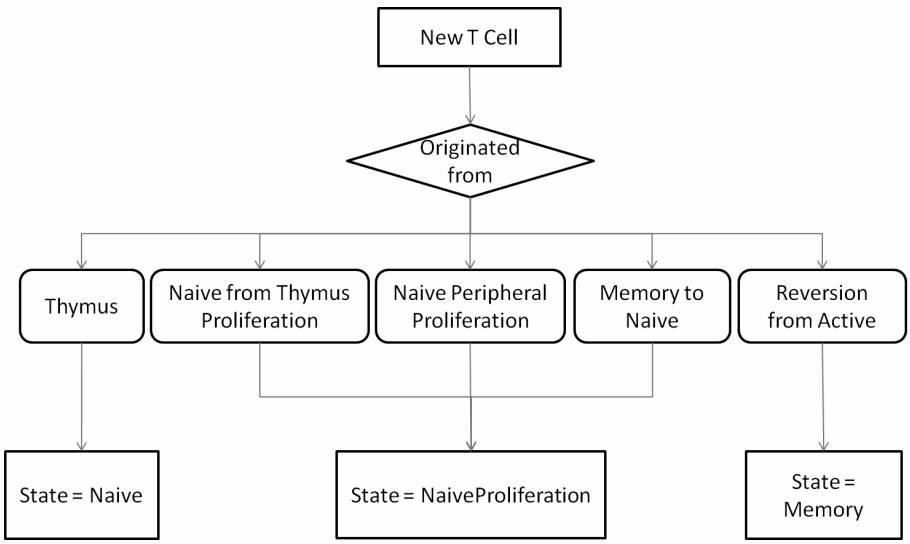


Figure 4.12: New agent (T cell) state decision flow chart

Simulation

For the simulation development, apart from the agents, there is also a function that determines the thymic output and the number of active cells (from the look-up table) that become memory cells. Both are implemented using events that determine when each of these T cells should enter the system. The thymic output calculation function and the active cells look-up table are the same as those from the SD model. Furthermore, as the ABMS model is built from the SD model, the functions s, g and h are also taken from the original SD.

4.2.6 Experiments

Five simulation scenarios were studied, defined by [59] with different values for the parameters. A summary of parameters changed for each scenario is illustrated in Table 4.5 below.

The first scenario investigated the need for naive peripheral proliferation throughout life. The naive peripheral proliferation rate for this experiment was therefore set to zero. It also considered reversion from memory to a naive phenotype.

Table 4.5: Simulation parameters for different scenarios. The parameter c is only used in the first scenario, where there is no proliferation. In the other scenarios, proliferation is defined by the equation $\left(1 + \frac{300}{\overline{N}_p}\right) * \frac{4.4}{h}$ [59].

Scenario	Description	Parameters						
		λ_n	λ_{mn}	$\overline{N}_p$	$\overline{s}$	b	μ_{N_p}	c
1	No peripheral proliferation	0.22	0.05	387	0.48	3.4	0.13	0
2	No homeostatic reduction in thymic export, no homeostatic alteration of naive death rate	2.1	0	713	0	0	4.4	–
3	Homeostatic alteration of naive death rate but not thymic export	0.003	0	392	0	4.2	4.4	–
4	Homeostatic alteration of thymic export but no naive death rate	0.005	0	378	2.4	0	4.4	–
5	No restrictions	0.005	0	378	2.2	0.13	4.4	–

The second scenario assumed peripheral proliferation with a higher rate of naive cells becoming naive proliferating cells ($\lambda_n = 2.1$). There was no reversion from memory to a naive phenotype and no homeostatic reduction in thymic export. The functions s, g and h from the mathematical model were responsible for controlling the thymic export, naive death rate and naive peripheral proliferation respectively. In order to alter the thymic export, the parameters $\overline{s}$ and $\overline{N}_p$ were changed. The parameter b was set to zero so that the function g would remain constant during the entire simulation, as would the death rate of naive cells.

The third scenario altered the function g over time by setting the parameter b greater than zero ($b = 4.2$). This meant that the death rate of naive T cells from thymus increased along the years as the number of naive from peripheral proliferations rose. There was no change to the thymic export, no reversion from memory to a naive phenotype and the conversion rate of naive from thymus to naive proliferation was low (equal to 0.003).

Scenario 4 produced the opposite results from those of scenario 3. In this case there was no change in the death rate of naive T cells from thymus. Rather, there was change on the thymic export with time.

Finally, the fifth scenario presented no restrictions, which meant that there were changes in thymic export and death of naive cells over time. Moreover, there was peripheral proliferation and no memory turning back to a naive phenotype.

The data used for validation of the simulations is displayed in Tables 4.6 and 4.7. The data set contains information about the TREC marker in individuals grouped in age ranges. The first column of Table 4.6 shows the age range of the individuals; the second column has the mean $\frac{\log_{10} TREC}{10^6} PBMC$ (peripheral blood mononuclear cell) and the third column contains the number of individuals in each age range. The total number of individuals in the experiment was 506.

The graphic containing the TREC data (naive from thymus) and total naive cell data provided by [59] [15] and [50] is shown in Figure 4.13. In the figure, data provided in Table 4.6 is represented by the symbol ◯ ; the □ symbol indicates the data from Table 4.7. In addition, in Figure 4.13 the total percentage of naive T cells in the body, obtained in [59], is also displayed (symbol ◊).

For the ABMS, the simulation was run fifty times and the mean result of these runs was collected.

Table 4.6: The data set used for validation obtained in [59] and [15]

Age	$\frac{\log_{10} TREC}{10^6 \times nPBMC}$	number of individuals
0	5.03	48
1-4	4.93	53
5-9	4.86	19
10-14	4.86	19
15-19	4.56	33
20-24	3.88	26
25-29	3.75	47
30-34	3.61	65
35-39	3.54	73
40-44	3.52	52
45-49	3.37	55
50-54	3.17	16

Each simulation was run for a period of one hundred years taking into account the impact of thymic shrinkage per mm^3 of peripheral blood and 3673 initial naive cells from thymus for the SD model: $10,000$ data points were collected during each run (one every 0.01 year).

Table 4.7: The data set collected in Lorenzi *et al.* [50]

Age	$\frac{\log_{10} TREC}{10^6 \times nPBMC}$	number of individuals
0	4.85	2
1-4	5.29	30
5-9	5.05	33
10-14	4.99	15
15-19	4.56	5
20-24	4.55	12
25-29	4.55	9
30-34	4.44	20
35-39	4.23	15
40-44	4.16	9
45-49	3.82	16
50-54	4.21	21

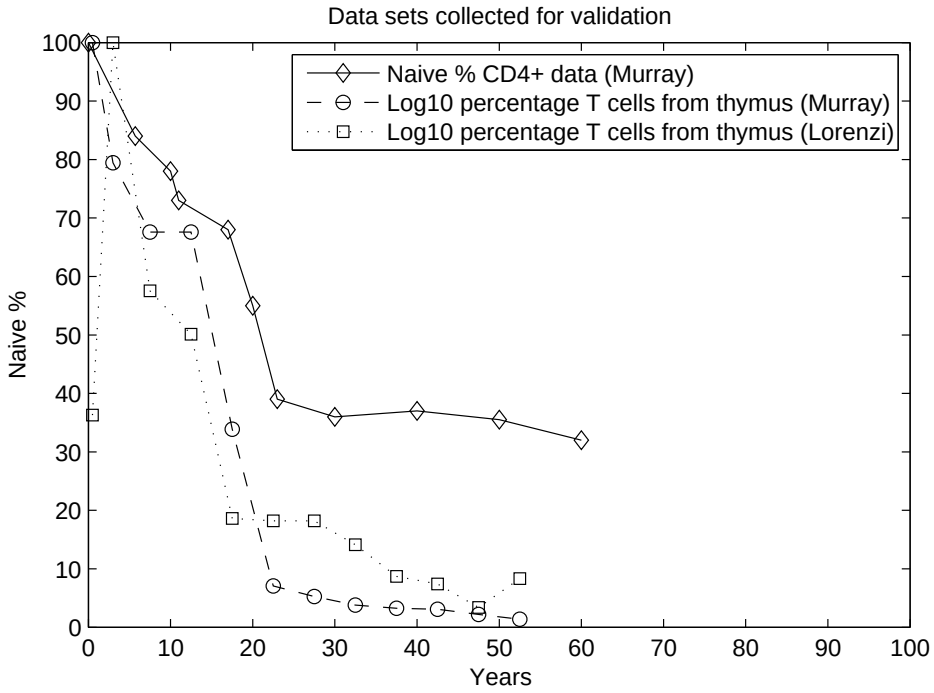


Figure 4.13: Data sets used for validation of the naive T cell output simulation models

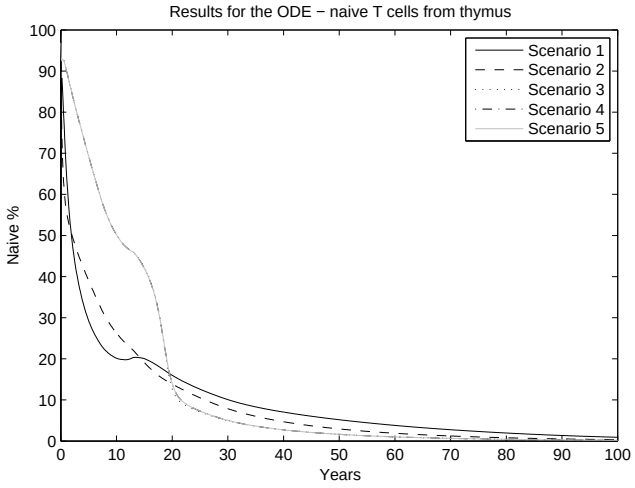
4.2.7 Results

The simulation results contrasting one SD run with one ABMS run are illustrated in Figures 4.14 , 4.15 and 4.16 below. Figures 4.14(a), 4.15(a) and 4.16(a) show the ODE results used as a baseline for our results validation. In the first scenario, the results for both simulation techniques show a very similar trend curve, although the ABMS results exhibit a more noisy behaviour in time. Results did not fit the original data (Tables 4.6 and 4.7). The resulting naive cells from thymus curve demonstrated a substantial decay in thymic export on the beginning of life because of the high death rate. In comparing SD and ABMS outputs, the results were similar. As expected, the ABMS simulation produced some variation on the simulation curves while SD's curve was steady. In addition, SD simulation took less computational resources.

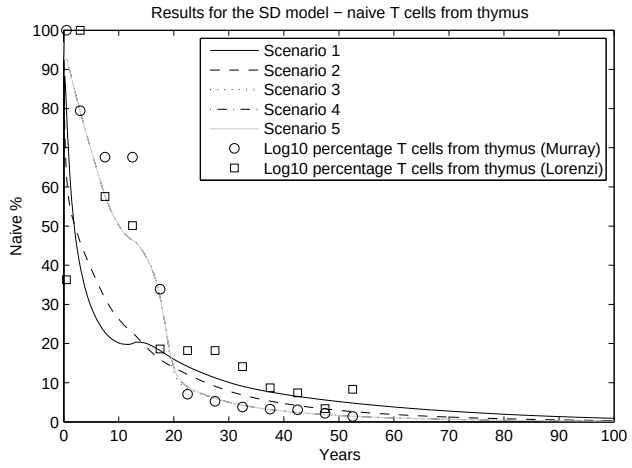
After the twenties, an exponential decay of thymic export was observed and the dynamics followed the thymic decay rate rule defined in the mathematical model. The naive proliferation curve increased with the decrease of naive from thymus, but as there was no proliferation of peripheral cells, they died with no replacement. Thus they followed the same pattern as that of their only source, i.e. thymic naive cells. The results indicate that peripheral proliferation is important for maintenance of naive T cells.

Results from scenario 2 matched the original data more closely. This case considered peripheral proliferation, as well as a high rate of naive cells from the thymus turning into peripheral naive cells. The naive from thymus curve shows a substantial decay in the beginning of life because of the death and proliferation rates. On the other hand, the naive from proliferation curve increased with the decrease of the naive from thymus curve. This pattern was controlled by the g function.

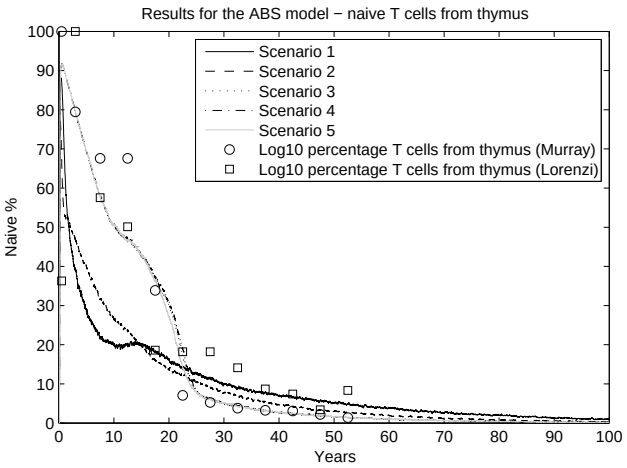
The main difference between these results and the results from the previous scenario is that the number of naive cells from proliferation reached a stable value after the age of twenty with no further decay. The results indicate the importance of peripheral expansion, but also the need for a smaller rate of naive to peripheral naive conversion. Moreover, reversion from memory to a naive phenotype is not important.



(a) ODE

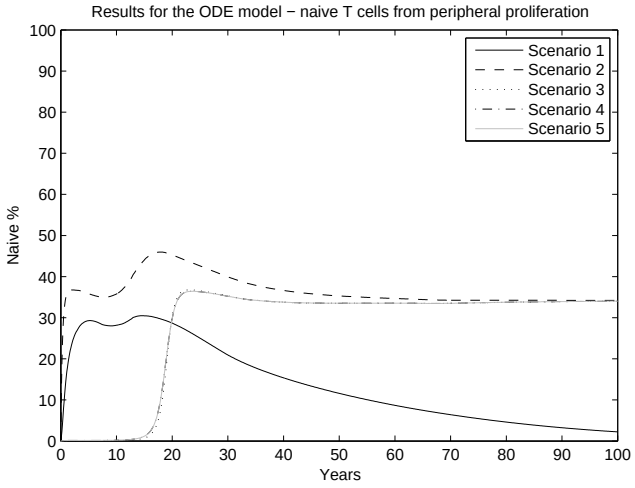


(b) SD

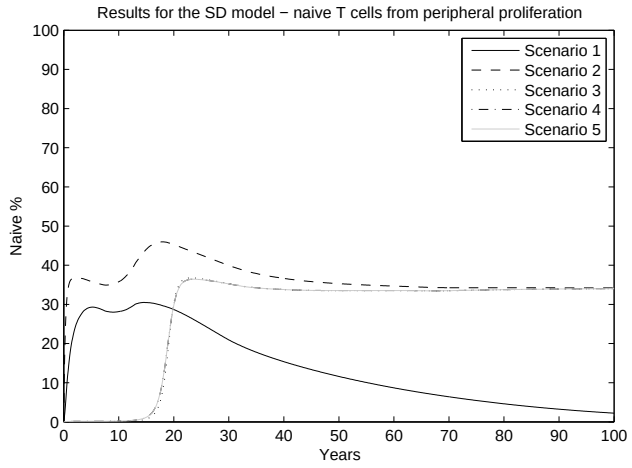


(c) ABMS

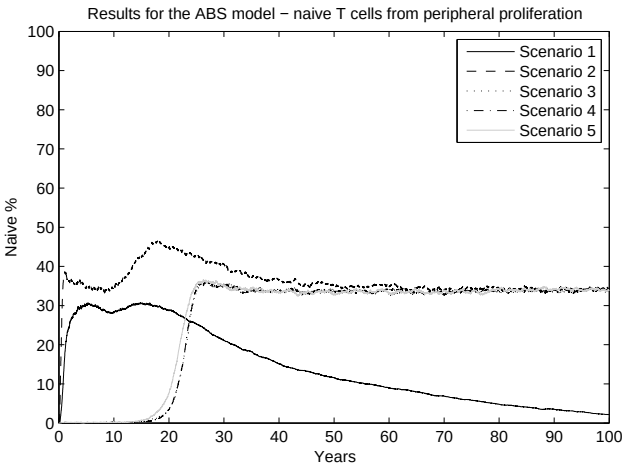
Figure 4.14: Results for naive T cells from thymus



(a) SD

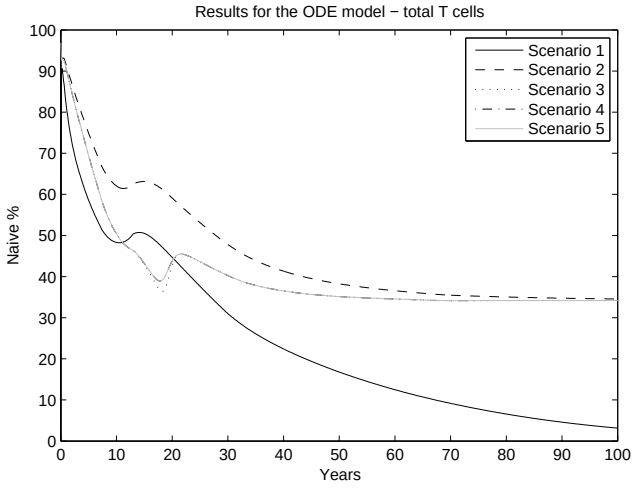


(b) SD

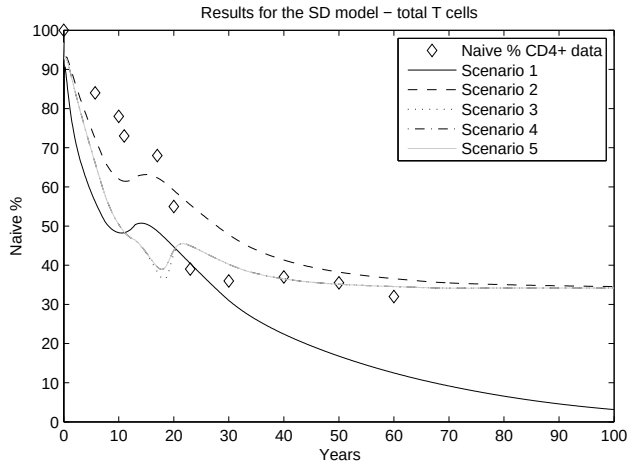


(c) ABMS

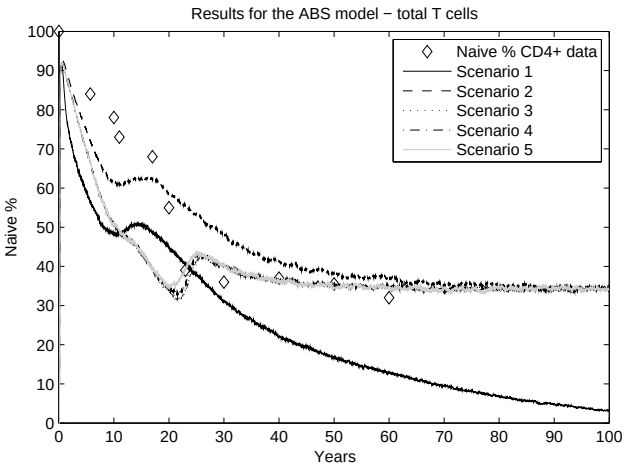
Figure 4.15: Results for naive T cells from peripheral proliferation



(a) ODE



(b) SD



(c) ABMS

Figure 4.16: Results for total T cells

Scenario 3 took into account the results produced in the previous scenarios and adjusted the parameters in a way that a more accurate output was obtained. The naive from thymus curve presented a decay at the beginning of life followed by an interval of stability. By the age of twenty the thymic export decreased in an exponential trend. With the decay of naive from thymus, the naive repertoire changed from the thymic source to peripheral proliferation source. By performing these simulations it is therefore possible to have an idea of how the decay of naive cells occurs over time. The results now closely matched the original data.

Scenarios 4 and 5 produced similar results to scenario 3. This indicates that alterations in thymic export and in naive death do not interfere significantly with the overall dynamics of the naive T cells.

In the five scenarios studied, the simulations produced similar results for both SD and ABMS. This can also be observed in the results of Wilcoxon rank sum tests applied to both ABMS and SD results for the simulations (Table 4.8). The table reports p-values associated with Wilcoxon rank sum tests for the five scenarios. Our hypothesis is that the outcomes produced are not significantly different. The p-values for each test all exceed the 0.05 (5%) significance level, indicating that the distributions of the outcomes of the various simulation approaches are not statistically different and therefore, the tests failed to reject the hypothesis.

Table 4.8: Wilcoxon test with 5% significance level comparing the results from SD and ABMS for the data sets used for validation on the number of naive T cells from thymus

Scenario	p
1	0.8650
2	0.8750
3	0.7987
4	0.8408
5	0.9719

4.2.8 Summary of Case 1

The main factors influencing the process of immunosenescence include the number and phenotypical variety of naive T cells in an individual, which change with age in quantity and diversity. At the beginning of life, the thymus is the principal source of naive T cells. With age, there is a decay in thymus output and a shift between the main source of naive T cells. It is believed that the sustenance of naive T cells in the organism is provided by peripheral expansion, reversion from a memory phenotype, and long-lived T cells.

The simulation models that were built and studied were based on mathematical equations converted into SD and from SD to ABMS. Five simulation scenarios were studied. The simulation outputs were broadly similar for both SD and ABMS. Our research question was to determine which of these two paradigms would be more suitable for the simulation of the static agents involved in this case study.

In the ABMS model, cells were subject to individual rates that occurred during the time slot in which they were created. This made the output for each run noisier than the SD results. It also seems to be a simulation process closer to reality, because in real immune systems, cells have individual behaviours and responses to the environment.

SD, on the other hand, gave a systemic view of the conceptual model and attempted to forecast how the system as a whole would evolve in time in an aggregate manner. This suggests that each change ratio would be applied to the entire set of cells. SD is simpler to implement and demands significantly less computational resources such as memory, processing time and complexity.

Results fit the observed data, and the likely contribution of each of the naive T cell repertoire maintenance method can therefore be estimated. With the decay of naive cells derived from the thymus, the naive repertoire changes from the thymic source to the peripheral proliferation source. The numbers of naive cells tend to be stable over time, but there is no new phenotypical naive cell entering the system.

The SD based simulation is closer to the underlying mathematics, but has the disadvantage of being high-level, with complete homogeneity of simulated entities. On the other hand, ABMS allows a representation of each entity and heterogeneity, although it increases the demand for computational resources.

4.3 Case 2: Tumour Growth

In the previous section, SD and ABMS were compared for a naive T cell output model, and it was concluded that for that case study SD is more suitable. There were a set of five scenarios in which the agents had no interactions and, for all scenarios, SD and ABMS produced similar outputs. SD is considered preferable, as it takes up less computational resources.

In order to continue our investigation, which considers the conversion between ODEs, SD and ABMS simulation approaches for non-interacting agents, mathematical models of general tumour growth have been explored. The choice for tumour growth models was due to the importance of immune research on the interactions between tumour cells and the immune system. Further in this thesis, case studies regarding these interactions are therefore considered (Chapter 5).

The main differences between this case study model and that from case 1 with regard to the experimentation phase are: (1) case 2 is a one-equation model, (2) the mathematical outcomes assume a "goal seeker" behaviour, where tumour cells increase or decrease according to parameter values, and (3) experiments with very small and very large population sizes are conducted.

4.3.1 Mathematical Models

This section presents the mathematical models used as a basis for the simulations. Tumour models involve only one equation, which defines mathematical rules for their growth. There are three classical models of tumour growth considered in this study: the logistics model, the von Bertalanffy model and the Gompertz model [21].

According to [21], the most general equation describing the dynamics of tumour growth can be written as:

$$\frac{dT}{dt} = Tf(T) \tag{4.7}$$

where:

- T is the tumour cell population at time t,
- $T(0) > 0$,
- $f(T)$ specifies the density dependence in proliferation and death of the tumour cells. The density dependence factor can be written as:

$$f(T) = p(T) - d(T) \tag{4.8}$$

where:

- $p(T)$ defines tumour cells proliferation
- $d(T)$ define tumour cells death

The expressions for $p(T)$ and $d(T)$ are generally defined by power laws:

$$p(T) = aT^{\alpha} \tag{4.9}$$

$$d(T) = bT^{\beta} \tag{4.10}$$

For our experiments, we defined the values for α and β using the three well-established models:

Logistics Model: $\alpha = 0$ and $\beta = 1$ ($a, b > 0$ and $b < a$ for growth) [21]

von Bertalanffy Model: $\alpha = \frac{-1}{3}$ and $\beta = 0$ ($a, b > 0$ and $b < a$ for growth) [21]

Gompertz Model: $p(T) = a$ and $d(x) = b\ln(T)$ ($a, b > 0$ and $e^b > a$ for growth) [21]

4.3.2 The System Dynamics Model

From Equations to System Dynamics

In order to obtain an equivalent SD model from the tumour growth equations, the guidelines defined in Section 3.5.1 have also been followed.

Model Stocks

For this model there is only one stock, which is the number of tumour cells.

Model Flows

The stock of tumour cells increases with the proliferation inflow and decreases with the death outflow, as shown in Figure 4.17:

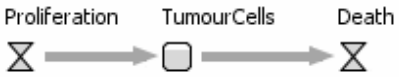


Figure 4.17: The tumour cells stock variable and its flows: proliferation and death

There is another way to implement the flows, which is considering

$$f(T) = Proliferation - Death$$

as an inflow that is positive for tumour growth or negative for tumour cells decrease (Figure 4.18):

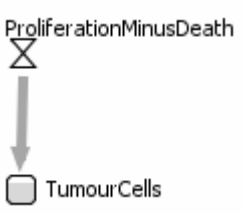


Figure 4.18: The tumour cells stock variable and its flow: $ProliferationMinusDeath$

We will use this second diagram in the remainder of this case study.

Model Information

Both proliferation and death occur according to the number of tumour cells, as defined in Equations 4.9 and 4.10. There is therefore information from the stock tumour cells to the $ProliferationMinusDeath$ flow, as shown in Figure 4.19:

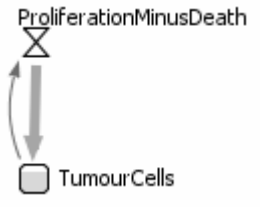


Figure 4.19: The tumour cells stock variable with its flow and information

Model Parameters

The model parameters are a, b, $alpha$ and $beta$ (Equations 4.9 and 4.10).

Flows Calculations

Table 4.9 shows how the equations from the mathematical models are defined for the flow values per each tumour growth model.

The Final System Dynamics Model

The final stock and flow diagram with information and parameters used for modelling the mathematical equations is shown in Figure 4.20.

Table 4.9: Flow values calculations

Model	ProliferationMinusDeath Flow
Logistic	$a \times TumourCells - b \times TumourCells^2$
von Bertanlaffy	$a \times TumourCells^{\frac{4}{3}} - b \times TumourCells$
Gompertz	$a \times TumourCells - b \times \ln(TumourCells) \times TumourCells$

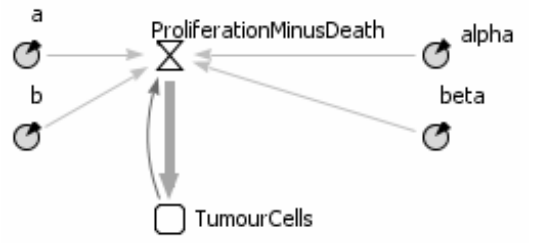


Figure 4.20: System dynamics model for the one-equation mathematical model

4.3.3 From System Dynamics to Agent-based Modelling and Simulation

In order to convert from SD to ABMS the guidelines introduced in Section 3.5.2 are once again used.

Model Agent

For the tumour growth case study, there is only one agent, the tumour cell (corresponding to the stock variable $TumourCells$ from the SD model stock and flow diagram).

Agent Behaviours

The tumour cell agent behaviours are "proliferate" or "die", according to the rate defined by the mathematical model. If the rate is positive, there is proliferation, otherwise, death occurs (Table 4.10). For the Gompertz model implementation, the parameters *alpha* and *beta* are not considered.

Table 4.10: Agents' parameters and behaviours for the tumour growth model

Parameters	Reactive behaviour	Proactive behaviour
a, *alpha*, b and *beta*	Dies if $rate < 0$	Proliferates if $rate > 0$

Agent Implementation

The tumour cell assumes two states, alive and dead, as shown in Figure 4.21 below. In the alive state, these cells can replicate and die. If the growth rate is positive, the cell replicates according to the rate value; otherwise, it dies. There is, therefore a branch connecting the two transitions $proliferate$ and *death* to the alive state. Once cells move to the final state dead, they are eliminated from the system and from the simulation.

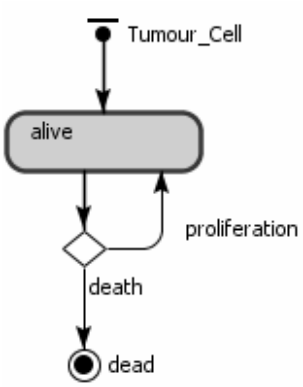


Figure 4.21: Tumour Cell agent

The transition connecting the state alive to the branch is triggered by the growth rate. The correspondent ABMS model rates to the flow values in the SD model are shown

in Table 4.11, which illustrates that the variable $TumourCellsAlive$ corresponds to the number of tumour cell agents in the state *alive* at a certain time in the simulation.

Table 4.11: Transition rate calculations from the SD flow equation for the tumour growth model

Model	SD Flow equation	Transition rate
Logistic	$a.TumourCells - b.TumourCells^2$	$a - (b.TumourCellsAlive)$
von Bertanlaffy	$a.TumourCells^{\frac{4}{3}} - b.TumourCells$	$a.TumourCellsAlive^{\frac{1}{3}} - b$
Gompertz	$aTumourCells - b\ln(TumourCells)TumourCells$	$a - b.\ln(TumourCellsAlive)$

Simulation

In the main simulation, apart from the tumour cell agents, the value for the parameters is defined according to the model studied and outcome values are saved.

4.3.4 Experiments

Two experiments were carried out to compare the SD and ABMS simulation outputs. For the first experiment, a variable c was established, representing the ratio between a and b. The purpose of c was to observe the impact of a and b on the tumour growth curve: c was therefore set as 5, 2.5, 1.7 and 1.25, so that a fair range of growth could be observed.

In the second experiment, we defined $a = 1.636$ and $b \in \{0.002, 0.005\}$. These values were determined in [45] and they were used for the next simulation set of experiments using a two-equation model (Chapter 5).

As the outcomes for ABMS were stochastic, each simulation was run for fifty times and the mean simulation output was presented. For both simulations, the initial values for tumour cells were defined equal to one.

4.3.5 Results

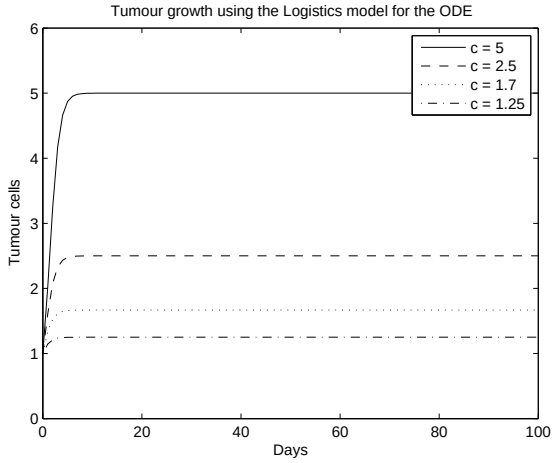
These experiments first validated our SD model by comparing its outputs with the outputs produced by the mathematical model derived from [21]. Both produced very similar results. Our ABMS model was then tested by comparing its outputs to the outputs produced by the SD model (i.e. our base model for the comparison).

The first experiment, as shown in Figures 4.22 (for the Logistics model), 4.23 (for the Von Bertanlaffy model) and 4.24 (for the Gompertz model) revealed that the outputs for both simulation approaches were similar for all models. In the logistic model the variance of the ABMS outcomes is higher because the number of cells in the simulation is small. In addition, the overall growth of cells in the ABMS results is more accentuated than the SD results. The Wilcoxon test shows therefore that the distributions of values for both outcomes differ at a 5% significance level for most scenarios (Table 4.12) (in the table we defined zero for the p-values which results were smaller than 10^{-6}). The test indicates similar results only for scenario 4 in the Logistic model and scenario 2 in the Von Bertalanffy model. This is explained by the discrete growth of agents in the ABMS model contrasted with the continuous growth of stocks in the SD model. Furthermore, the SD is deterministic, while the ABMS is stochastic. In this regard, the outcomes of the ABMS has erratic behaviour, as is shown in Figure 4.22 (b).

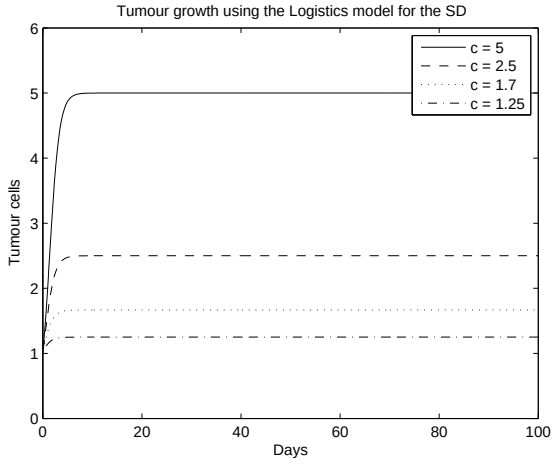
Table 4.12: Wilcoxon test with 5% significance level comparing the results from SD and ABMS for the Tumour Growth model. The hypothesis of similarity for most cases was rejected ($p \simeq 0$)

Scenario	p-value (Logistic)	p-value (Von Bertanlaffy)	p-value (Gompertz)
1	0	0	0
2	0	0.4859	0
3	0	0	0
4	0.3221	0	0

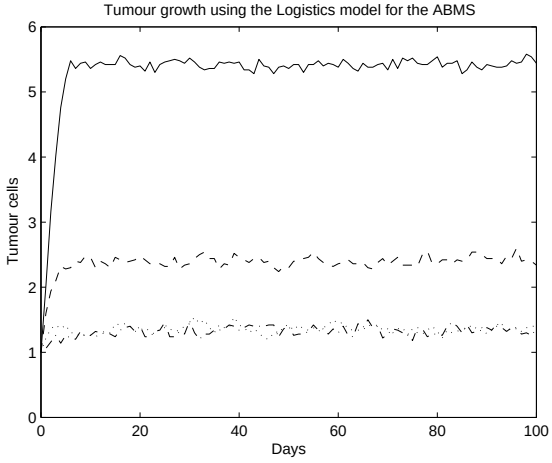
Figure 4.25 shows the results for the second experiment, which are also similar, although Wilcoxon test results reject the similarity hypothesis. The literature suggests that the logistics model is one of the most used for average tumours, whereas the von Bertalanffy and Gompertz models are used for more aggressive tumours. As the difference between a



(a) ODE

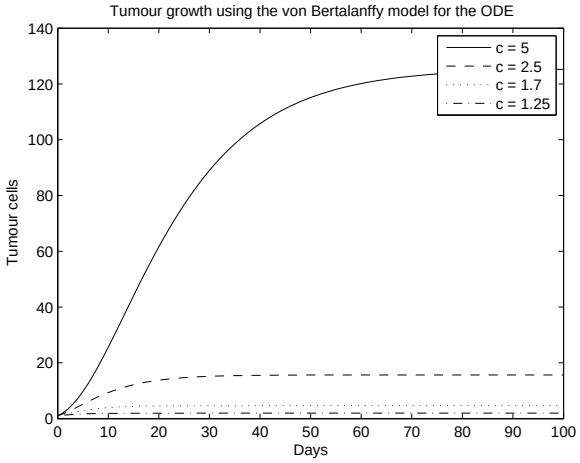


(b) SD

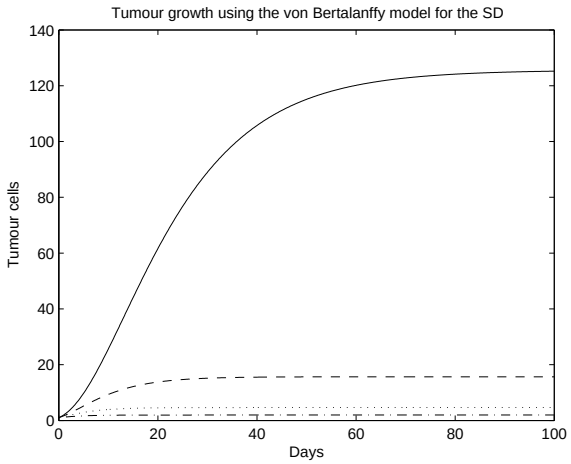


(c) ABMS

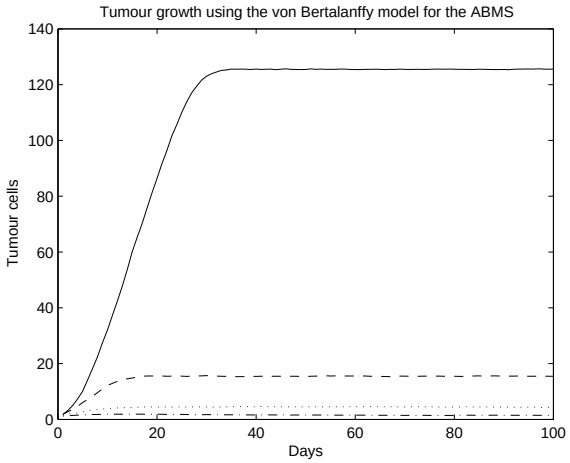
Figure 4.22: Results for the tumour growth model varying the c parameter: Logistic model



(a) ODE

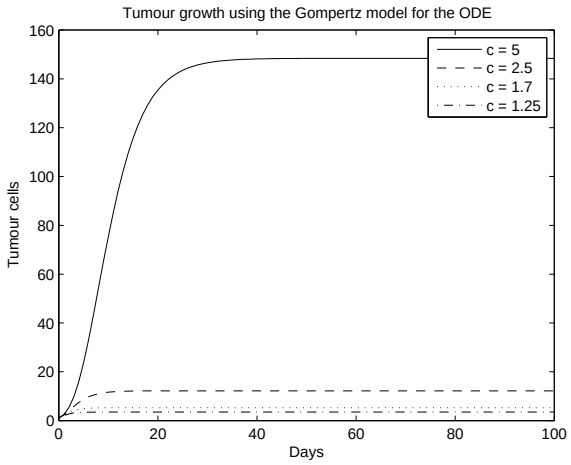


(b) SD

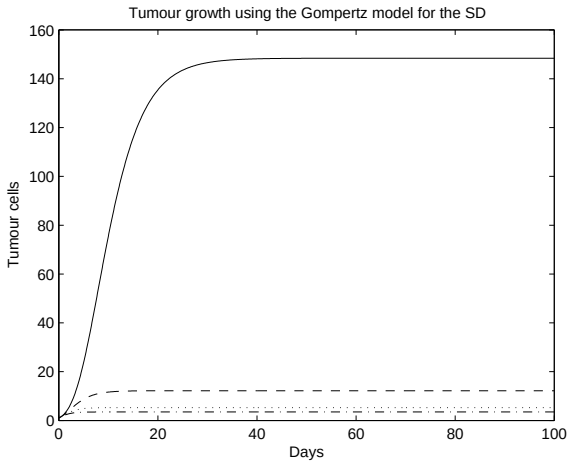


(c) ABMS

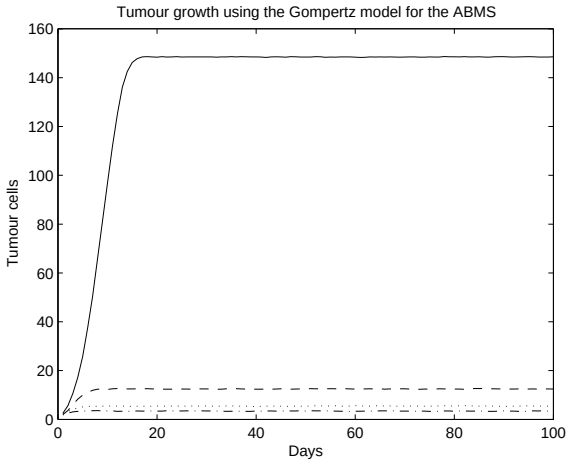
Figure 4.23: Results for the tumour growth model varying the c parameter: Von Bertalanffy model



(a) ODE



(b) SD



(c) ABMS

Figure 4.24: Results for the tumour growth model varying the c parameter: Gompertz model

and b is increased, the growth in the Gompertz and von Bertalanffy models demonstrates a considerable increase on the proliferation of tumour cells, as illustrated in Figure 4.26 below.

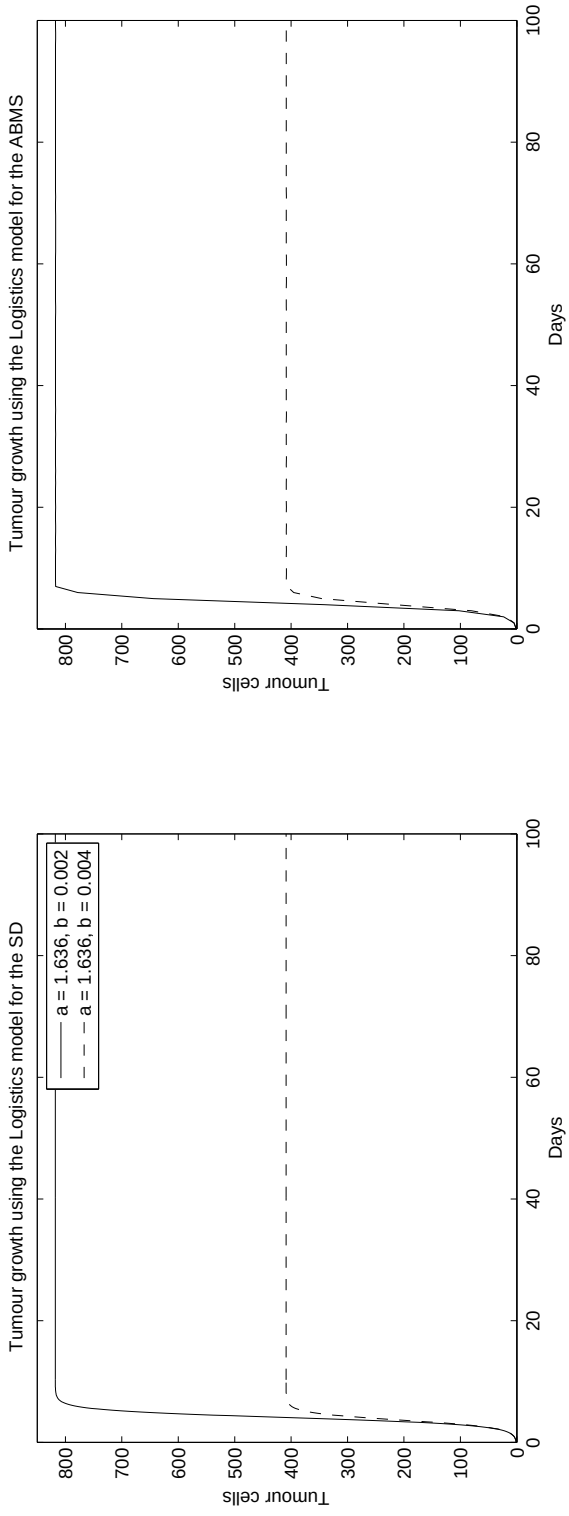
Consequently, experiment two using ABMS for the Gompertz and von Bertalanffy models could not be conducted. From the SD results, it can be observed that the number of tumour cells reaches over 10^{64} in the Gompertz model, depicted in Figure 4.26(a). To run the same experiment with ABMS more computational resources are required, and it would take up more processing time. In this case it is therefore preferable to run the simulation using SD, even though such a high number of tumour cells also seems to be unrealistic in tumour biology.

4.3.6 Summary of Case 2

In the second case study simulations from mathematical models of tumour growth were built. The models used were reviewed in [21] and represented three different tumour growth patterns, namely, the logistics growth model, the von Bertalanffy model and the Gompertz model. These models were used to test our conversion guidelines and to compare SD and ABMS outputs. The mathematical models were converted into SD and the ABMS model was developed from the SD model. The intention was to check if the results would be similar and if SD and ABMS could be used interchangeably for the second case study.

Two experiments were conducted to compare the outputs for the models. In the first experiment, the outputs for both simulation approaches were similar. However, the initial growth of the ABMS results is more accentuated given the fact that agents are discrete values. For the second experiment, considering a larger number of tumour cells, the results were very similar.

It was also observed that the SD simulation was more suitable when the number of tumour cells increased considerably, especially those models of aggressive tumours (von Bertanlaffy and Gompertz).



(d) SD

(e) ABMS

Figure 4.25: Results for the second experiment of one-equation logistics model using SD and ABMS

(a) Von Bertalanffy

(b) Gompertz

Figure 4.26: Results for the second experiment of tumour growth Von Bertalanffy (a) and Gompertz (b) models using SD

4.4 Case 3: Viral Spread of Human Immunodeficiency Virus

The third case study tests the conversion guidelines and compares SD and ABMS results for an ODE model of cell-free viral spread of human immunodeficiency virus (HIV) in the bloodstream [17]. The model consists of three populations: healthy CD4+ T Cells, infected CD4+ T cells and the concentration of free HIV at a certain time. The model objective is to observe the dynamics of these populations over time.

Healthy CD4+ cells are susceptible to HIV infection at any time during the progress of the disease. Any cell that expresses CD4 receptors in its surface can be infected. In particular, CD4+ T cells are widely attacked. CD4+ T cells, also known as T helper cells, are responsible for coordinating most of the adaptive immune responses. AIDS (acquired immune deficiency syndrome) is the disease of the human immune system caused by the loss of T cell populations, which are compromised by the HIV. This condition progressively reduces the immune system effectiveness and leaves the organism susceptible to opportunistic infections and tumours.

A T cell invaded by a HIV assumes two states. The first is *infected*, in which the HIV penetrates the cell. Once the HIV infects the cell, it starts replicating itself inside the cell. Subsequently, an infected cell becomes *actively infected*. When reaching this state, the lytic death of the T cell occurs, which releases the free HIV replications into the organism, increasing the infection rate. Further details regarding the interactions between T cells and HIVs are introduced in the next section.

4.4.1 Mathematical Model

The mathematical model proposed in [17] is described by equations 4.11 to 4.13. In these equations, T is the concentration of healthy CD4+ T cells, I is the concentration of infected CD4+ T cells, V is the concentration of free HIV and t is time (in days). Healthy CD4+ T cells can be infected by HIV according to a certain rate. Once the

infection occurs, the HIV is no longer part of the free HIV concentration.

Equation 4.11 describes the dynamics of healthy T CD4+ cells over time. These cells are (1) produced by precursor cells, (2) die with age, (3) grow by proliferation and according to the number of infected CD4+ cells and (4) get infected according to the number of healthy CD4+ cells and the amount of free viruses in the organism. Equation 4.12 describes the dynamics of actively infected cells, as they are obtained by infection of free HIV within a healthy CD4+ T cell and die according to a certain rate. The concentration of free HIV dynamics is described by equation 4.13, in which they are produced whenever an infected cell dies and releases more free viruses; these free viruses infects new cells and die according to a certain rate.

There is a percentage of infected cells that becomes actively infected; however, in the model, only actively infected cells are considered. The three mathematical equations are depicted as follows:

$$\frac{dT}{dt} = s - \mu_T T + rT\left(1 - \frac{T+I}{T_{Max}}\right) - k_1 VT \tag{4.11}$$

$$\frac{dI}{dt} = k_1' VT - \mu_I I \tag{4.12}$$

$$\frac{dV}{dt} = n\mu_b I - k_1 VT - \mu_V V \tag{4.13}$$

where:

- $T(t)$ represents the concentration of healthy CD4+ T cells at time t
- $I(t)$ represents the concentration of infected CD4+ T cells at time t
- $V(t)$ represents the concentration of free HIV at time t
- s is the source of CD4+ T cells from precursors
- μ_T is the natural death rate of CD4+ T cells
- r is the growth rate of CD4+ T cells

- T_{Max} is CD4+ T cells carrying capacity
- k_1 represents the rate of infection of T-cells with free virus
- k_1' is the rate at which infected cells become actively infected
- μ_I is the death rate of infected cells
- n represents the source for free virus
- μ_b is the lytic death rate for infected cells
- μ_V is the loss rate of virus

4.4.2 The System Dynamics Model

From Ordinary Differential Equations to System Dynamics

In this section, the ODE model introduced in Section 4.4.1 is converted into an SD model.

Model Stocks

Following the guidelines from Section 3.5.1, there are three stock variables: the healthy CD4+ T cells ($HealthyCD4$), the infected CD4+ T cells ($InfectedCD4$) and the free HIV concentration ($FreeHIVConcentration$).

Model Flows

The $HealthyCD4$ stock inflows are source newborn cells ($Source$) and cellular growth ($Growth$). The outflows are cellular death ($HealthyCD4Death$) and infection ($Infection$), as shown in Figure 4.27.

The $InfectedCD4$ stock is changed by source (infected cells that become actively infected) and death (Figure 4.28).

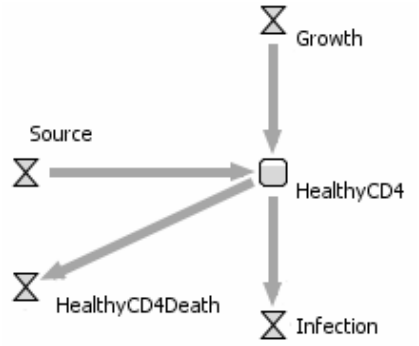


Figure 4.27: The healthy CD4 cells stock and its flows: source (*Source*), cellular growth *Growth*), death (*HealthyCD4Death*) and infection (*Infection*)

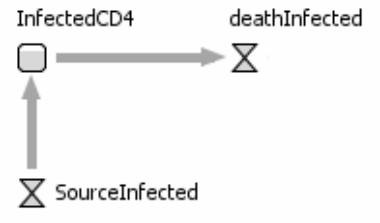


Figure 4.28: The infected CD4 cells stock and its flows: source (*SourceInfected*) and death (*deathInfected*)

The *FreeHIVConcentration* stock changes with source, infection of CD4+ cells and death of viruses (Figure 4.29).

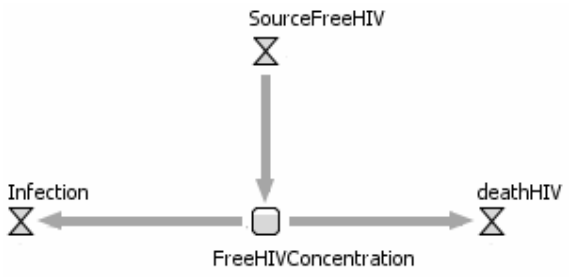


Figure 4.29: The free HIV stock and its flows: source (*SourceFreeHIV*), infection of CD+ cells (*Infection*) and viral death (*deathHIV*)

Model Information

By looking at Equation 4.11, it is possible to identify information between the stock $HealthyCD4$ and the flows $Growth$, $Infection$ and $HealthyCD4Death$, as shown in Figure 4.30. In Equation 4.11, for the growth calculation, there is the need for information coming from infected cells, illustrated in the complete SD stock and flow diagram (Figure 4.33).

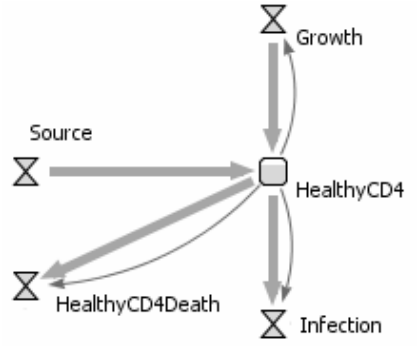


Figure 4.30: The $HealthyCD4$ stock with its flows and information

Equation 4.12 shows information from $InfectedCD4$ to $deathInfected$. As previously stated, there is also information from infected cells to the flow $Growth$ (Figure 4.31).

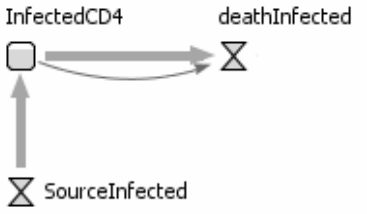


Figure 4.31: The $InfectedCD4$ stock with its flows and information

There is information from the $FreeHIVConcentration$ stock to the flows $Infection$ and $deathHIV$ (Equation 4.13), as shown in Figure 4.32.

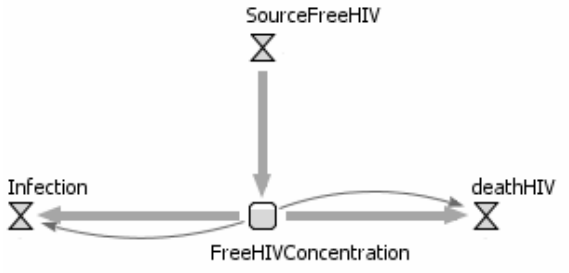


Figure 4.32: The *FreeHIVConcentration* stock with its flows and information

Model Parameters

The model parameters are the same as those from the mathematical model. Table 4.13 below depicts the mathematical parameters, their equivalents in the SD model and their values for the simulation. These values were defined in [17].

Table 4.13: Parameters from the mathematical model, their correspondents and values for the case 3 SD model

Rate	Correspondent	Value
s	$SourceCD4$	10
μ_T	$deathRateHealthyCD4$	0.02
r	$growthRate$	0.03
T_{Max}	$CarryingCapacity$	1500
k_1	$infectionRate$	0.000024
k_1'	$becomeActivelyInfected$	0.00002
μ_I	$deathRateInfectedCD4$	0.26
n	$viralParticlesReleased$	500
μ_b	$lyticDeathRateInfectedCells$	0.24
μ_V	$deathRateHIV$	2.4

Flows Calculations

Table 4.14 shows the flows for each stock, their correspondent in the mathematical model and the flow formula.

Table 4.14: Flow calculations for the viral spread model

Stock	Flow	Expression	Flow formula
HealthyCD4	*Source*	s	$SourceCD4$
	Growth	$rT\left(1-\frac{T+I}{T_{Max}}\right)$	$growthRate.HealthyCD4.\left(1-\frac{HealthyCD4+InfectedCD4}{CarryingCapacity}\right)$
	Infection	k_1VT	$(infectionRate.HealthyCD4\ FreeHIVConcentration)$
	HealthyCD4Death	$\mu_T T$	$(deathRateHealthyCD4.\ HealthyCD4)$
InfectedCD4	*SourceInfected*	$k_1'VT$	$(becomeActivelyInfected.\ FreeHIVConcentration.\ HealthyCD4)$
	DeathInfected	$\mu_I I$	$deathRateInfectedCD4.\ InfectedCD4$
FreeHIVConcentration	*SourceFreeHIV*	$n\mu_b I$	$(viralParticlesReleased.\ lyticDeathRateInfectedCells.\ InfectedCD4)$
	DeathFreeHIV	$\mu_V V$	$(deathRateHIV.\ FreeHIVConcentration)$

The Final System Dynamics Model

All the stocks combined with the flows and parameters defined in the mathematical model form the SD model and can be seen in Figure 4.33.

4.4.3 From System Dynamics to Agent-based Modelling and Simulation

Model Agents

In order to define the agents the first guideline defined in Section 3.5.2 is reviewed:

- stocks preferably become states when they represent accumulations of elements from the same population
- stocks become agents when they represent accumulations from different popula-

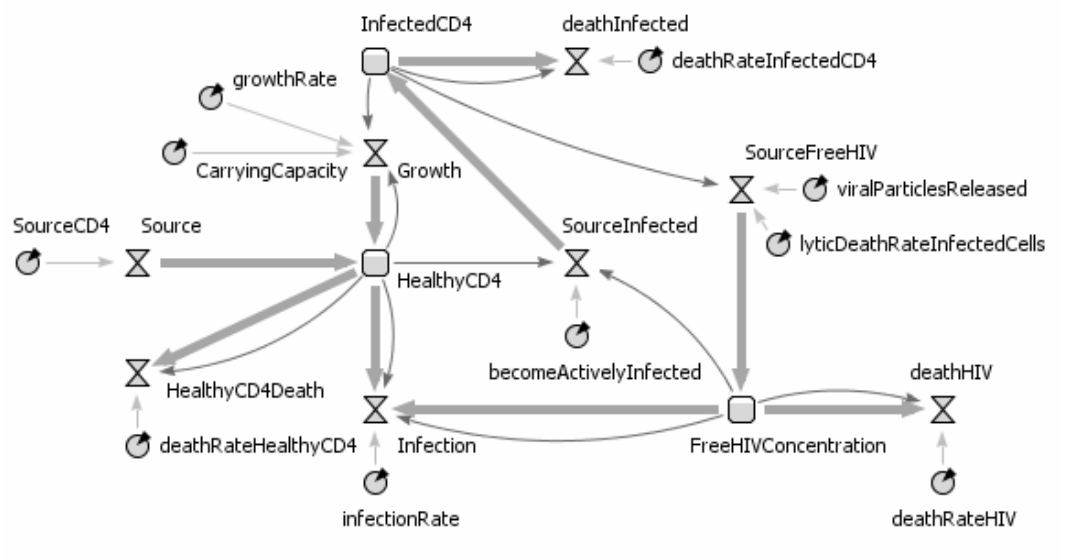


Figure 4.33: The system dynamics model's functions and parameters for case 3

tions

In case 3, the CD4+ T cell population is initially healthy and then becomes infected and actively infected, according to predefined rates (Table 4.13). As the stocks $HealthyCD4$ and $InfectedCD4$ represent accumulations of the same population, they are therefore included in the state chart diagram for the ABMS model as states of the T cell CD4 population of agents. For the T cell agent, therefore, we have the states $Healthy$ and $ActivelyInfected$.

The other agent in the system is the free HIV virus. This agent has only one intermediate state, when it is alive and free in the system. It can also die or infect a cell (final states). Both final states reduce the free HIV agents population.

Agents' Behaviours

The agents' parameters and behaviours corresponding to each state are shown in Table 4.15 below.

Table 4.15: Agents' parameters and behaviours for the viral spread model

Agent	State	Parameters	Reactive behaviour	Proactive behaviour
T Cell	*Healthy*	$deathRateHealthyCD4$	Dies	–
		$becomeActivelyInfected$	Become infected	–
		$growthRate$ and $CarringCapacity$		Grows
	ActivelyInfected	$deathRateInfectedCD4$ and $viralParticlesReleased$	Dies	–
		$lyticDeathRateInfectedCells$	HIV Concentration	–
HIV	*Alive*	$infectionRate$	Infects Healthy CD4	–
		$deathRateHIV$	Dies	–

Agents' Implementation

Figure 4.34 depicts the state chart diagram for the CD4+ T cell agents, in which CD4+ T cells agents are created in the initial state $HealthCD4$, where they also grow (reproduce). These agents change state (either death or $ActivelyInfected$) according to the rates defined in the mathematical model. When in the state $ActivelyInfected$, T cell agents release a number of free HIVs in the system.

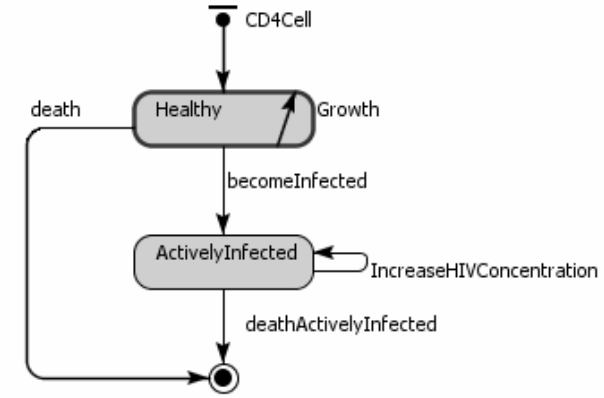


Figure 4.34: The CD4+ T cell agent state chart

The other agent in the system is the free HIV virus. For the SD, the concentration of virus is considered. In the ABMS, however, due to the fact that the number of agents is a discrete number, each virus is considered as an agent. As shown in Figure 4.35 below, the free HIV agent dies and infects cells according to the respective rates. When the

infection transition is triggered, the HIV agent sends a message to the CD4 Cell agent so that it becomes infected. Furthermore, during the infection, the HIV agent is removed from the free HIV population.

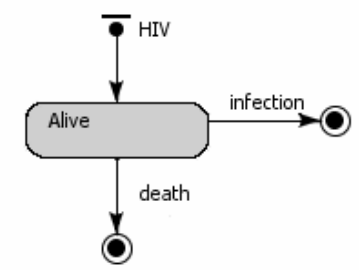


Figure 4.35: The free HIV agent state chart

The ABMS model rates corresponding to the flow values in the SD model are shown in Table 4.16, in which the variable $TotalHIV$ corresponds to the number of free HIV agents in the system at a certain time in the simulation. Similarly, the variable $TotalHealthy$ is the total number of healthy T cells and $TotalInfected$ is the total number of actively infected cells.

Table 4.16: Transition rates calculations from SD flows equations for the viral spread model

State	Transition	SD Flow equation	Transition rate
healthy (CD4)	becomeActively-Infected	($becomeActivelyInfected$. $FreeHIVConcentration$. $HealthyCD4$)	triggered by message
	death	($deathRateHealthyCD4$. $HealthyCD4$)	$deathRateHealthyCD4$
	growth	$growthRate.HealthyCD4$. $\left(1-\frac{HealthyCD4+InfectedCD4}{CarryingCapacity}\right)$	$growthRate.TotalHealthy$. $\left(1-\frac{(TotalHealthy+TotalInfected)}{CarryingCapacity}\right)$
Actively Infected (CD4)	deathActively-Infected	($deathRateInfectedCD4$. $InfectedCD4$)	$deathRateInfectedCD4$
Alive (HIV)	infection	($infectionRate$. $FreeHIVConcentration$. $HealthyCD4$)	$infectionRate.TotalHealthy$
	death	($deathRateHIV$. $FreeHIVConcentration$)	$deathRateHIV$

Simulation

For the simulation development, apart from the agents, there are also have three events, which are implemented methods in the Any Logic environment that occur according to a certain rate. The event *Source* determines the input of new T cells (which corresponds to the flow *Source* in the SD stock and flow diagram of Figure 4.33). This event recurrently occurs at a rate equal to 10 and adds a new healthy T cell in the system.

4.4.4 Experiments

For case 3, experiments were conducted initially using the parameter values defined in Table 4.13. Results were validated using those obtained in the mathematical model from [17]. As the outcomes for ABMS are stochastic, each simulation was run for fifty times and the mean simulation output was presented.

4.4.5 Results

The SD and ABMS simulation outcomes represented the dynamics of healthy CD4+ T cells, actively infected T cells and free HIVs. Experiments were carried out using exactly the same parameters and initial values for both simulations, but the results were very different. It was acknowledged that the initial value for free HIVs in the SD model was less than one, which could not be adopted in the ABMS as the number of agents was always an integer value; the initial HIV number was therefore increased to one in the SD. The SD results remained very similar as those of the mathematical model. The same value adopted in the ABMS, however, produced very different results. The HIV population disappears over the simulation and the infection is not established. There was the need, therefore to add a constraint in the free HIV agent, which determines that the number of agents should never be smaller than one, as shown in Figure 4.36. The branch in the state chart only allows viral death if the size of the population is greater than one.

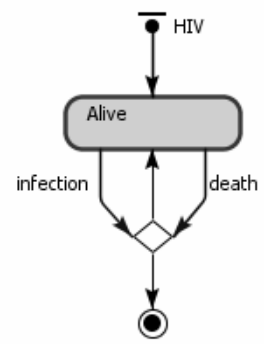
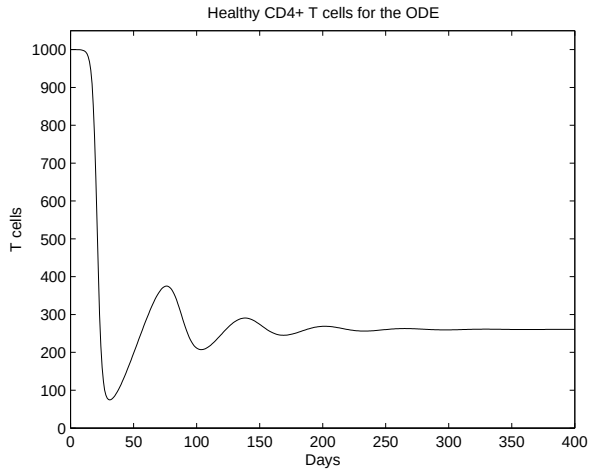


Figure 4.36: The free HIV agent state chart with constraint

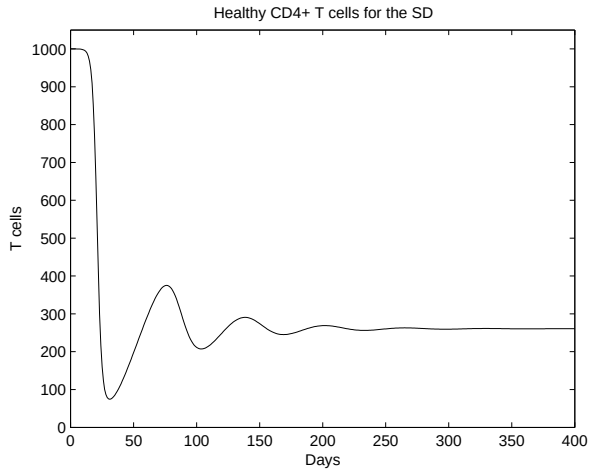
The simulation outcomes with the new HIV state chart are shown in Figure 4.37 (for healthy CD4+ T cells), Figure 4.38 (for infected CD4+ T cells) and Figure 4.39 for free HIVs. For the ABMS results, we show the outcomes of ten different runs. For each run, healthy CD4 starts decreasing at a different point in time. The decay of these cells is given by the HIV infection, which is defined by the rate $infectionRate \times TotalHealthy$. The healthy decrease start time will correspond to any point in time delimited by the probability density function $f = e^{(infectionRate \times TotalHealthy)}$, which establishes the randomness of the system and consequently explains the differences occurred between the ten runs in the graph.

The results with the closest ABMS run to the SD results are shown in Figures 4.40, 4.41 and 4.42. Although in this case the outcomes are more similar, there are still differences given by random character of the ABMS.

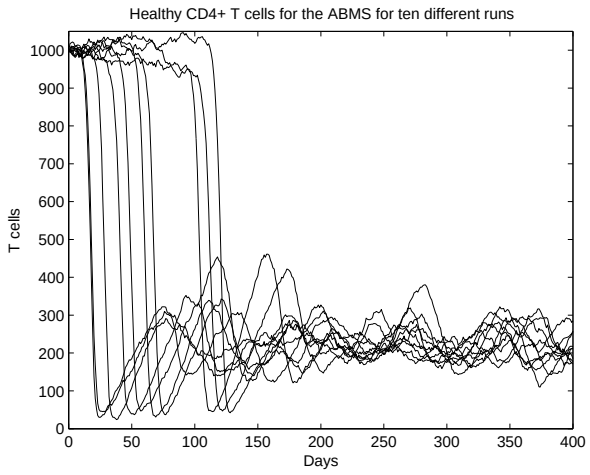
Furthermore, the numbers if actively infected cells and HIV in the ABMS are higher on the global local maximus. This occurs because of the continuous values considered in the SD model contrasted with the discrete number of agents from the ABMS.



(a) ODE

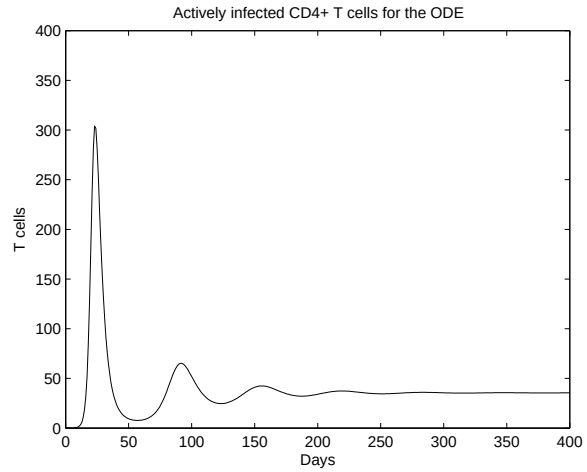


(b) SD

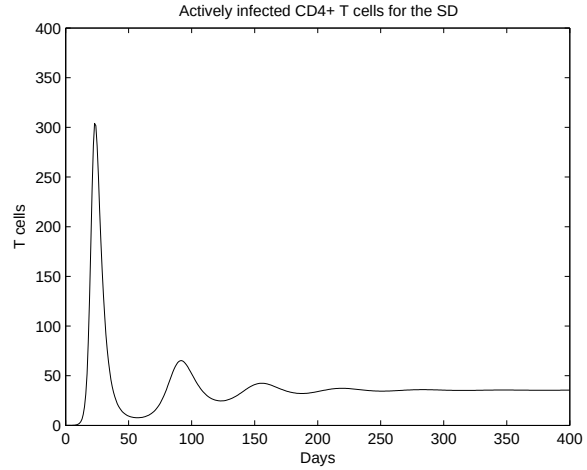


(c) ABMS

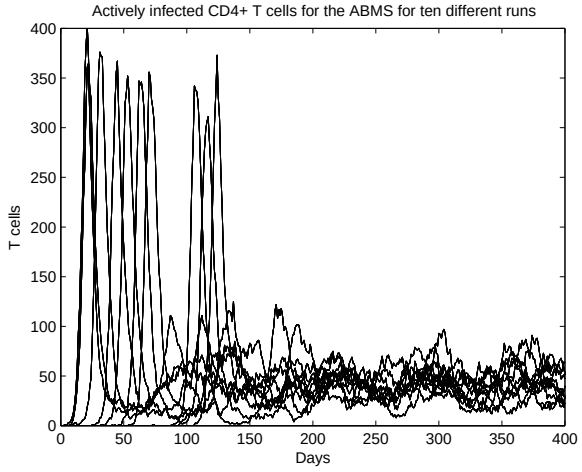
Figure 4.37: ODE, SD and ABMS results for the healthy CD4+ T cells



(a) ODE

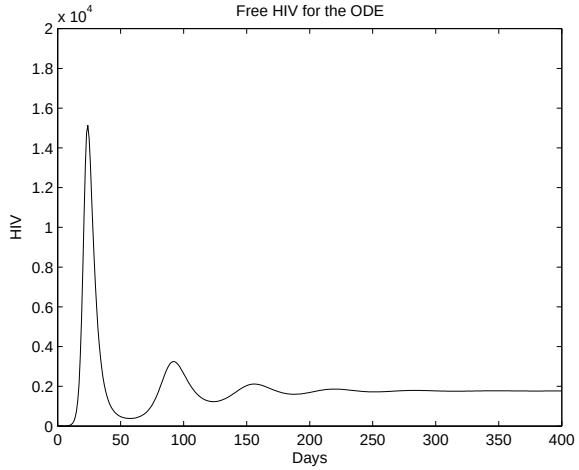


(b) SD

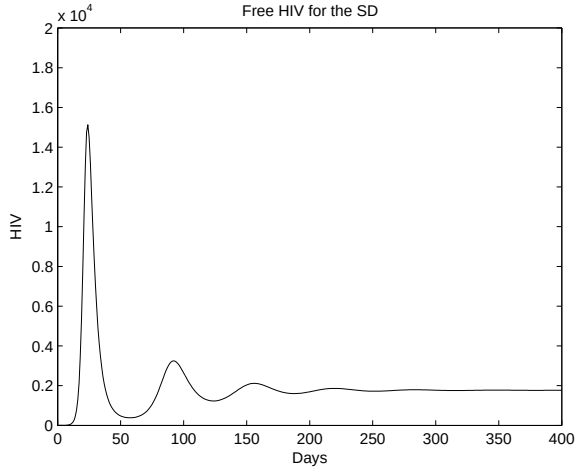


(c) ABMS

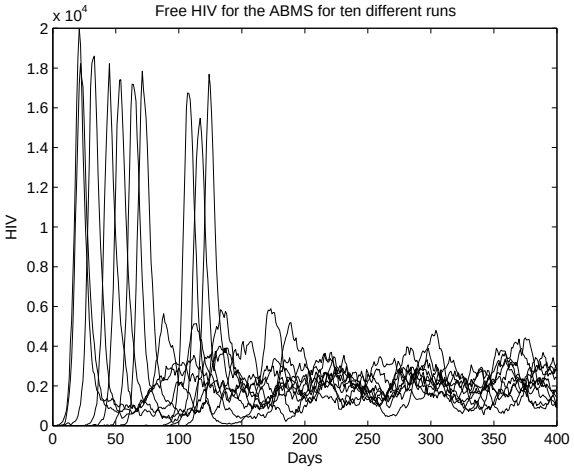
Figure 4.38: ODE, SD and ABMS results for the actively infected CD4+ T cells



(a) ODE

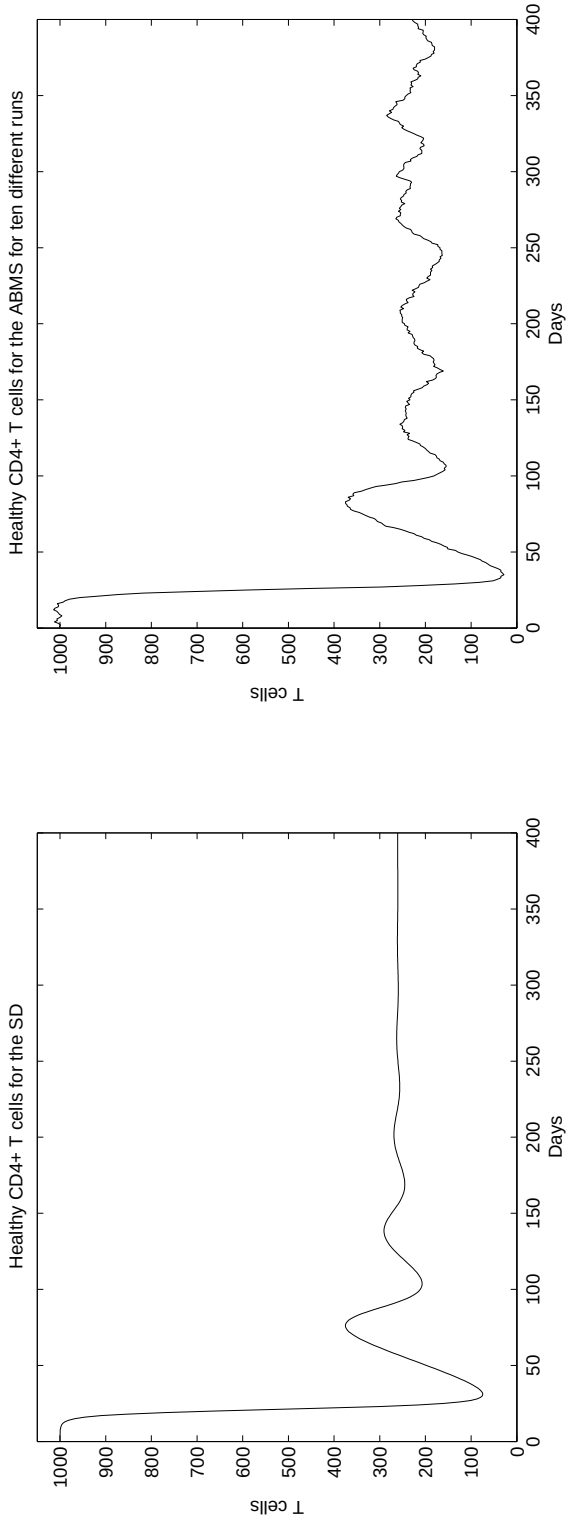


(b) SD



(c) ABMS

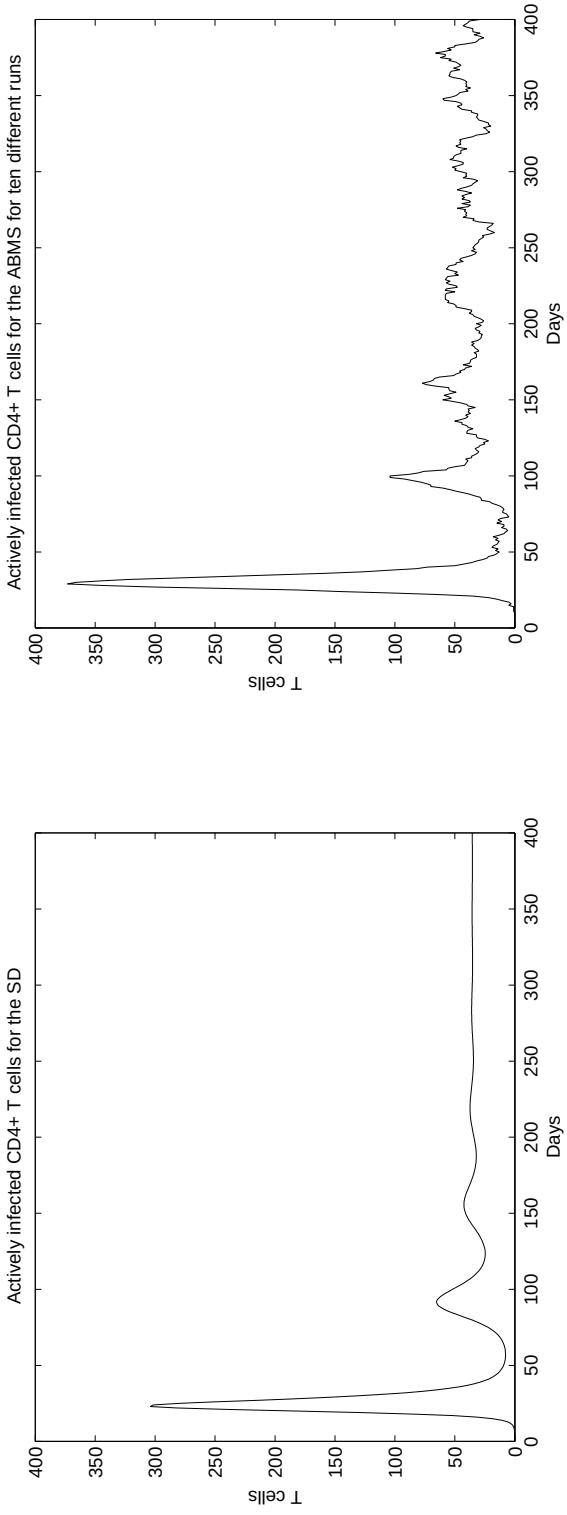
Figure 4.39: ODE, SD and ABMS results for the HIVs



(d) SD

(e) ABMS

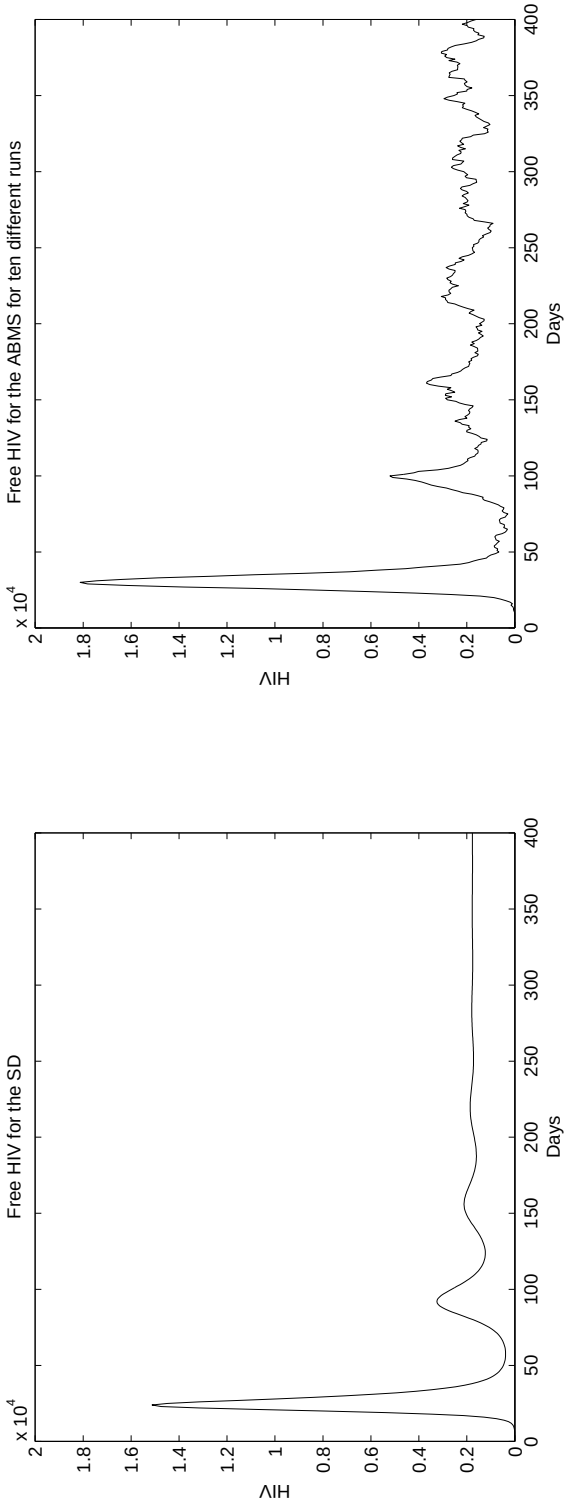
Figure 4.40: SD and ABMS closest results for the healthy CD4+ T cells



(a) SD

(b) ABMS

Figure 4.41: SD and ABMS closest results for the actively infected CD4+ T cells



(a) SD

(b) ABMS

Figure 4.42: SD and ABMS closest results for the HIVs

4.4.6 Summary of Case 3

In the third case study our conversion guidelines were tested and SD and ABMS results for a HIV spread mathematical model were compared. The mathematical equations describe the dynamics of populations of healthy CD4 T cells, infected CD4 T cells and free HIV viruses. With the conversion guidelines we developed, it was possible to build the SD and ABMS simulations. The SD results matched exactly the mathematical model outcomes (see SD results from Figures 4.37, 4.38 and 4.39), but the results for the ABMS were different.

The differences between the SD and ABMS results were due to the fact that stocks in the SD assumed real numbers while agents were integer values. Furthermore, the stochasticity inherent in the ABMS model produced several different runs with distinct times in which the HIV infection started. In all the runs, however, the pattern of behaviour, that is the decrease of healthy cells followed by an increase of actively infected and free HIV virus, occurs in all the runs. And in this sense, the ABMS results seem more realistic, as an infection of an organism can occur or develop at any point whithin a time interval.

4.5 Summary

This chapter presented three case studies with different characteristics in order to explore the effectiveness of our conversion guidelines and asses the different approaches results after the conversions. Furthermore, we wanted to investigate the most suitable simulation approach for examples involving static non-spatial entities.

The first case study was concerned with the use of ODEs to model interactions between different populations of T cell. The objective was to investigate the relevance of different sources of naive T cells to the organism. Case one was the most complex in terms of elements involved, populations size, modelling effort and time demanded for the simulations. The conversion steps allowed for the construction of the SD and the ABMS

without any extra effort. Regarding the outcomes, the Wilcoxon rank sum statistical test failed to reject the hypothesis that the outcomes for the SD and ABMS have equally large values. The main difference observed for both approaches concerned processing time and computational resources demanded: ABMS was far more time consuming and demanded more computational memory to run the experiments. For this case, it can be therefore concluded that, although the approaches can be used interchangeably, SD is more suitable.

The second case study referred to the investigation of tumour growth, and it was the most simple of the three cases presented. It was similar to the first example in that the conversion guidelines were effective in assisting the construction of both SD and ABMS simulations. It was observed that the ABMS presented limitations in simulating cases of aggressive tumours. As tumour cells increased significantly and very quickly, the ABMS runs were very slow and, in most of the cases, they exceed the available memory limit. As a result, only SD was effective in producing outcomes for these cases. Furthermore, it was observed that the overall growth for the ABMS outcomes is more accentuated than those from the SD, given the fact that tumour cells agents are discrete numbers.

In case three, although the conversion guidelines also made it possible to build both simulations, the results initially were very different. The original mathematical model investigated the dynamics of T Cells and HIVs in infected organisms. The first challenge encountered during the ABMS experimentation phase was that the HIV agents disappeared from the population and did not come back. There was the need, therefore, to introduce a constraint in the model to assure that at least one HIV agent was present throughout the simulation time. In addition, the main differences after adding the constraint were caused by the continuous real values for stocks against discrete number of agents.

By observing the case studies outcomes, it is not possible to establish a generalization as to when each approach is preferable. As a "rule of thumb" from our case studies, however, it seems that:

- When the population sizes increase to the order of 10^2, the outcomes from ABMS become very similar to those from the SD.
- In population sizes of this magnitude, therefore SD is preferable, as it is less resource consuming and simpler to implement.
- SD is also preferable for these cases because it is simpler to verify and validate against the ODE outcomes.
- ABMS is incapable to fully reflect the ODE outcomes when the mathematical model considers values smaller than one. Further validation with real data, therefore is necessary to evaluate which model better matches reality.

In the following chapter another set of case studies is presented which involves interacting agents. The effectiveness of our conversion guidelines is also assessed, and comparison of results is produced.

Chapter 5

Dynamic Spatial and Non-Spatial Models

5.1 Introduction

In this chapter a further three case studies involving simulations in immunology are developed in order to fulfil some of the objectives introduced in Section 3.3. In particular, we consider interacting non-spatial and spatial entities in the models. We believe that spatial movement will provide means to further test our conversion guidelines. We also hypothesise that spatial interactions between the elements of the models are more suitable for the ABMS models and their conversion to an equivalent SD might result in a rather complex stock and flow diagram. The goals of this chapter, therefore are:

1. To test the guidelines defined to convert from ODE to SD and to convert between SD and ABMS models and assess the impact of this conversion.
2. To compare SD and ABMS outcomes, the type of hypothesis to be tested and the modelling effort.
3. To define guidance to choose between SD and ABMS depending on the characteristics of the problem to be addressed.

As discussed in the previous chapter, simulations are built taking into account three different case studies based on mathematical modelling, in which we test the conversion guidelines, compare SD and ABMS outcomes and assess the impact of the conversions. The case studies are based on different non-spatial ODE models of interactions between the immune system and cancer. For the three case studies, the SD model is firstly obtained and converted into an ABMS model. Subsequently, an ABMS model with spatial interactions is derived from the non-spatial model. We investigate then if the spatial ABMS can be converted into into an SD model.

The fourth case study is based on an ODE model involving interactions between tumour cells and generic effector cells (Section 5.2). The fifth case study adds to the previous model the influence of IL-2 cytokine molecules in the immune responses of effector cells towards tumour cells (Section 5.3). The final case study comprises an ODE model of interactions between effector cells, tumour cells, and IL-2 and TGF-β molecules (Section 5.4). In Section 5.5, we discuss the results obtained and draw conclusions on the most suitable approach for each case, based on the outcomes obtained with the experiments.

5.2 Case 4: Interactions between Tumour Cells and Generic Effector Cells

In the previous chapter (Chapter 4), for the second case study, different types of mathematical models of tumour growth were converted into SD and ABMS simulation. For case 4 in this chapter, a mathematical model of tumour cells growth and their interactions with general immune effector cells defined in [45] is considered for the conversions into SD and ABMS.

Effector cells are responsible for killing the tumour cells inside the organism. Their proliferation rate is proportional to the number of tumour cells in the organism. As the quantities of effector cells increase, the capacity of eliminating tumour cells is boosted.

Furthermore, these immune cells proliferate and die per apoptosis, which is a programmed cellular death. Moreover, in the model, cancer treatment is also considered. The treatment consists of injections of new effector cells into the organism. The details of the mathematical model are given in the following section.

5.2.1 The Mathematical Model

The interactions between tumour cells and immune effector cells can be defined by the following equations:

$$\frac{dT}{dt} = Tf(T) - d_T(T, E) \tag{5.1}$$

$$\frac{dE}{dt} = p_E(T, E) - d_E(T, E) - a_E(E) + \Phi(T) \tag{5.2}$$

where

- T is the number of tumour cells,
- E is the number of effector cells,
- $f(T)$ is the growth of tumour cells,
- $d_T(T, E)$ is the number of tumour cells killed by effector cells,
- $p_E(T, E)$ is the proliferation of effector cells,
- $d_E(T, E)$ is the death of effector cells when fighting tumour cells,
- $a_E(E)$ is the death (apoptosis) of effector cells,
- $\Phi(T)$ is the treatment or influx of effector cells.

Kuznetsov [45] defines the functions $f(T)$, $d_T(T, E)$, $p_E(E, T)$, $d_E(E, T)$, $a_E(E)$ and $\Phi(t)$ as shown below:

$$f(T) = a(1 - bT) \tag{5.3}$$

$$d_T(T, E) = nTE \tag{5.4}$$

$$p_E(E, T) = \frac{pTE}{g + T} \tag{5.5}$$

$$d_E(E, T) = mTE \tag{5.6}$$

$$a_E(E) = dE \tag{5.7}$$

$$\Phi(t) = s \tag{5.8}$$

where:

- a and b are parameters for the Logistic growth of tumour cells and determine how fast the growth occur,
- n is the rate in which effector cells kill tumour cells,
- The parameters p and g control the amount of proliferation for the effector cells based on the number of existing tumour cells,
- m is the rate of death of effector cells after fighting tumour cells,
- d is the rate of effector cells apoptosis,
- s is the rate of influx of effector cells as treatment

The Logistic model is adopted for tumour growth, as it seems to be the most commonly used among the mathematical models involving cancer and the immune system.

5.2.2 The System Dynamics Model

From Ordinary Differential Equations to System Dynamics

As in the previous chapter, the guidelines of Section 3.5.1 to obtain the SD model are applied.

Model Stocks

The stocks for case 4 are the tumour cells and the effector cells.

Model Flows

The tumour cells stock is modified by proliferation and death of these cells, which in Figure 5.1 below are combined in only one flow $ProliferationMinusDeath$, which is an inflow to represent the tumour growth. The stock is also modified by the number of tumour cells that are killed by the effector cells:

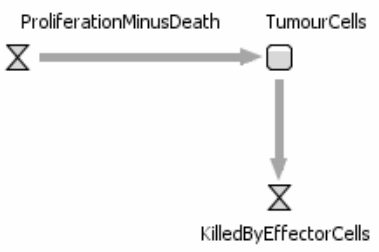


Figure 5.1: The tumour cells stock variable and its flows: $proliferationMinusdeath$ and tumour cells $killedbyEffectorCells$

The effector cells stock is modified by the inflows proliferation and treatment; and by the outflows death and apoptosis, as shown in Figure 5.2:

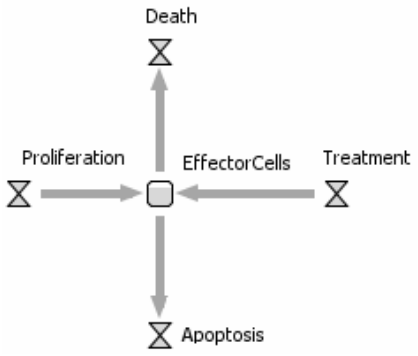


Figure 5.2: The effector cells stock variable and its flows: proliferation, death, apoptosis and treatment

Model Information

There is information from the $TumourCells$ stock to $ProliferationMinusDeath$ and $KilledByImmuneCells$ flows, as shown in Figure 5.3. In addition, the number of tumour cells influences the death of effector cells and also stimulates the proliferation of these immune cells. This information is included in the complete SD model shown in Figure 5.5.

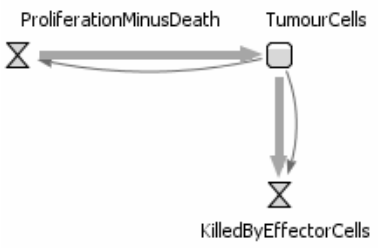


Figure 5.3: The tumour cells stock variable with its flows and information

There is information from the $EffectorCells$ stock to the flows $Death$, $Proliferation$ and $Apoptosis$, as shown in Figure 5.4. In addition, the number of effector cells influences the number of tumour cells killed (Figure 5.5).

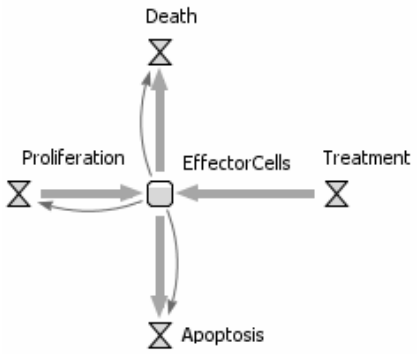


Figure 5.4: The effector cells stock variable with its flows and information

Model Parameters

The model parameters are the same as those from the mathematical model: a, b, d, g, m, n, p and s. The values for these parameters vary according to the scenario investigated (Section 5.2.4).

Flows Calculations

Table 5.1 shows how the equations from the mathematical models are defined for the flow values:

The Final System Dynamics Model

The complete SD is shown in Figure 5.5.

5.2.3 From System Dynamics to Agent-based Modelling and Simulation

Model Agents

Two classes of agents are defined: the tumour cell and the effector cell.

Table 5.1: Flow calculations for case 4

Stock	Flow	Expression	Flow formula
TumourCells	$ProliferationMinusDeath$	$a(1-bT)T$	$TumourCells\times$ $(a \times (1 - b \times TumourCells))$
	$KilledByEffectorCells$	nTE	$n \times TumourCells \times EffectorCells$
EffectorCells	$Proliferation$	$\frac{pTE}{g+T}$	$\frac{p\times TumourCells\times EffectorCells}{g+TumourCells}$
	$Death$	mTE	$m \times TumourCells \times EffectorCells$
	$Apoptosis$	dE	$d \times EffectorCells$
	$Treatment$	s	s

Agents' Behaviours

The agents' parameters and behaviours corresponding to each state are shown in Table 5.2 below. In this example, all behaviours are derived from the flows in the SD stock and flow diagram, so no extra modelling effort is necessary apart from those indicated in our guidelines:

Table 5.2: Agents' parameters and behaviours for case 4

Agent	Parameters	Reactive behaviour	Proactive behaviour
Tumour Cell	a and b	Dies (with age)	
	a and b		Proliferates
	m		Damages effector cells
	n	Dies killed by effector cells	
Effector Cell	m	Dies (with age)	
	d	Dies per apoptosis	
	p and g		Proliferates
	s	Is injected as treatment	

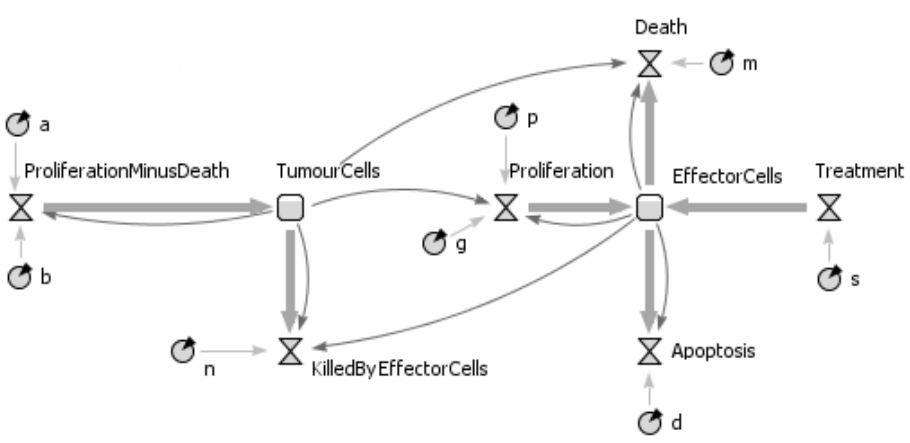


Figure 5.5: SD model for case 4

Agent Implementation

The state chart for the tumour cells is shown in Figure 5.6(a), in which an agent proliferates, dies with age or is killed by effector cells. In addition, at a certain rate, the tumour cells contribute to damage to effector cells. The rates defined in the transitions are the same as those from the mathematical model. Figure 5.6(b) presents the effector cell agent state chart, in which either the cell is alive and able to kill tumour cells and proliferate or is dead by age or apoptosis.

(a) Tumour cell agent

(b) Effector cell agent

Figure 5.6: ABMS state charts for case 4

The ABMS model rates corresponding to the flow values in the SD model are shown in Table 5.3. In the transition rate calculations, the variable $TotalTumourCells$ corresponds to the total number of tumour cell agents; and the variable $TotalEffectorCells$

is the total number of effector cell agents:

Table 5.3: Transition rates calculations from SD flows equations for case 4

Agent	Transition	SD Flow equation	Transition rate
Tumour Cell	proliferation	$a.TumourCells.(1 - TumourCells.b)$	$a - (TotalTumour.b)$
	death	$a.TumourCells.(1 - TumourCells.b)$	$a - (TotalTumour.b)$
	dieKilledByEffectorCells	$n.TumourCells.EffectorCells$	$n.TotalEffectorCells$
	causeEffectorDamage	$m.TumourCells.EffectorCells$	m
Effector Cell	Proliferation	$\frac{p.TumourCells.EffectorCells}{g+TumourCells}$	$\frac{p.TotalTumourCells}{g+TotalTumourCells}$
	DieWithAge	$EffectorCells.d$	d
	DiePerApoptosis	$m.TumourCells.EffectorCells$	message from tumour

Simulation

For the simulation building, apart from the agents, there is also an event – namely, treatment – which includes new effector cells with a rate defined by the parameter s.

5.2.4 Experiments

Four experiments for case 4 were carried out and their parameter variation is shown in Table 5.4. These values were obtained from [21]. In the four scenarios, differences in the death rate of tumour cells (defined by parameter b), effector cells apoptosis rate (defined by parameter d) and treatment (parameter s) were considered. In the first three scenarios, cancer treatment was considered, while the fourth case did not consider any treatment. Similar to the experiments reported in the previous chapter, the simulation for the ABMS was run fifty times and the mean values are displayed as results.

Scenario	b	d	s
1	0.002	0.1908	0.318
2	0.004	2	0.318
3	0.002	0.3743	0.1181
4	0.002	0.3743	0

Table 5.4: Simulation parameters for different scenarios. For the other parameters, the values are the same in all experiments, i.e. $a = 1.636$, $g = 20.19$, $m = 0.00311$, $n = 1$ and $p = 1.131$, as defined in [45]. The four scenarios investigate variations in the aggressiveness of the tumour (parameters a and b), apoptosis of effector cells (parameter d) and amount of cells injected as treatment (s). p and g define the proliferation of effector cells and n determines the rate in which effector cells kill tumour cells.

5.2.5 Results

In the first scenario results shown in Figure 5.7 below, the behaviour of the tumour cells appears similar for ODE, SD and ABMS. However, the Wilcoxon test rejected the similarity hypothesis for tumour outcomes, as shown in Table 5.5. The reason for this test pointing out that the outcomes differ is that tumour cells for the ODE and SD models decreased asymptotically towards to zero, while the ABMS behaviour is discrete and therefore **reached zero**. Furthermore, the variances observed in the ABMS curve, given its stochastic characteristic, also influenced the Wilcoxon test results. The number of effector cells for both simulations follow the same pattern, although the numbers are not the same due to the agents variability. This variability is very evident with regards to the effector cells population for two main reasons: (1) for this case study the size of the populations involved is relatively small, which increases the impacts of stochasticity in the outcomes; and (2) the ODE and SD systems change the amount of cells overtime in a continuous fashion, which means that, in this simulation, fractions of cells are considered. ABMS does not consider fraction of cells - a cell either is alive or dead. This is implemented as a boolean indicator and corresponds to the real world, where fractions of cells could obviously not exist. Considering the above explanations we conclude that for this scenario the ABMS outcomes seem more realistic, as in biological experiments cells are also atomic entities and stochastic variability occurs.

The results for the second scenario seem similar for effector cells, as shown in Figure 5.8, which was confirmed by the Wilcoxon test (Table 5.5). The results for the tumour cells are visibly not the same. Regarding the ODE and SD simulations, in the first ten days the tumour cells population first decreases and then grows up to a value of 240 cells, in which the growth reaches a steady-state. The initial decrease of tumour cells is also observed in the ABMS outcomes. After ten days, however, there is a smaller cellular increase and a steady-state is not observed. Similar to the previous scenario, the simulation curve presents an erratic behaviour throughout the simulation days. There is, however an unexpected decay of tumour cells over time. This is explained by the individual characteristics of the agents and their growth/death rates attributed to their instantiation. As the death rates of tumour cells agents are defined according to the mathematical model, when the tumour cell population grows, the newborn tumour cells have higher death probabilities, which leads to a considerable number of cells dying out. This indicates that the individual behaviour of cells can lead to a more chaotic behaviour when compared to the aggregate view observed in the ODE and SD simulations.

Table 5.5: Wilcoxon test comparing case 4 simulation results

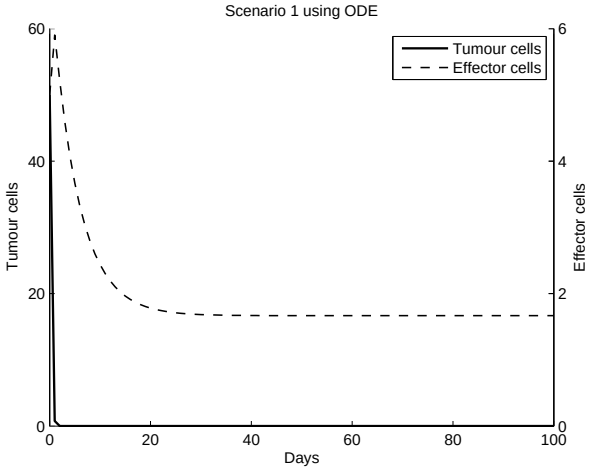
Implementation	**Cells**	**Scenario** (p-value)			
		1	2	3	4
ABS	*Tumour*	0	0	0.8591	0
	Effector	0.3789	0.6475	0	0
ABS - Fix 1	*Tumour*	0	0	0	0.0011
	Effector	0	0.3023	0	0
ABS - Fix 2	*Tumour*	0	0	0	0
	Effector	0	0	0	0

For scenarios 3 and 4, shown in Figures 5.9 and 5.10 respectively, the results for both approaches differ completely. Moreover, with regard to the tumour cells curve, the differences are even more evident. The ODEs and SD outcomes for scenario 3 reveal that tumour cells decreased as effector cells increased, following a predator-prey trend curve. For the ABMS, however, the number of effector cells decreased until a value close to zero was reached, while the tumour cells numbers varied differently from the ODEs results. As we discussed for the previous scenarios, the predator prey-pattern observed

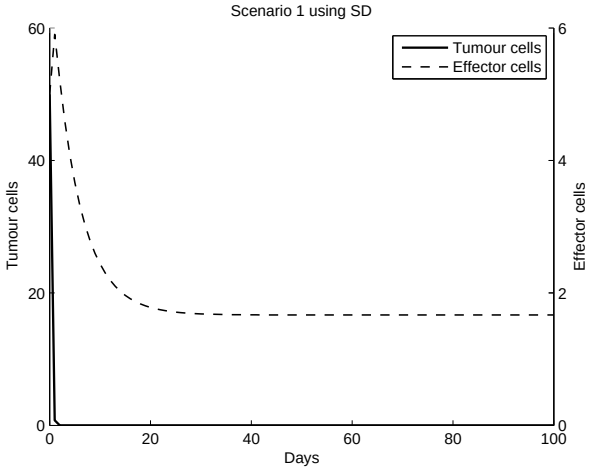
in the ODE and SD simulations was only possible due to its continuous character. In the ODE and SD simulation outcome curves for the effector cells it is possible to observe, for instance, that after sixty days the number of effector cells ranges between one and two. These values could not be reflected in the ABMS simulation and therefore the differences occur.

In scenario 4, although effector cells appear to decay in a similar trend for both approaches, the results for tumour cells vary widely. In the SD simulation, the numbers of tumour cells reached a value close to zero after twenty days and then increased again. For the ABMS simulation, however, tumour cells reached zero and never increased again. Similar to scenarios 2 and 3, the continuous ODE simulation outcomes contrasted with discrete agents caused the different outcomes. Furthermore, as occurred in scenario 2, the individual behaviour and rates attributed to the cells seemed to have an impact in the growth of tumours.

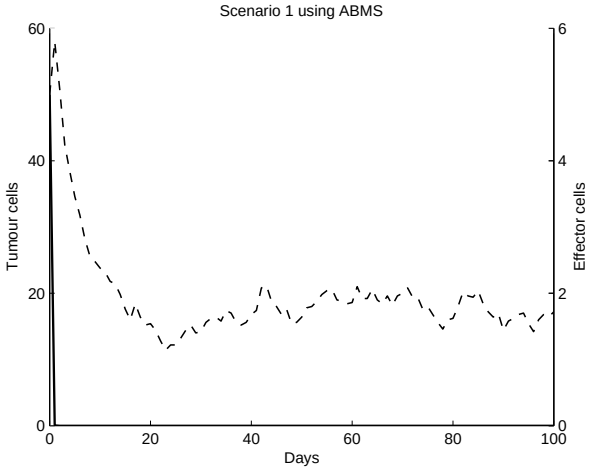
Overall, regarding the processing time taken and computational resources used to run the simulations, both approaches were equivalent. As there were only a few agents in the system, the ABMS simulations ran nearly as fast as those using SD.



(a) ODE

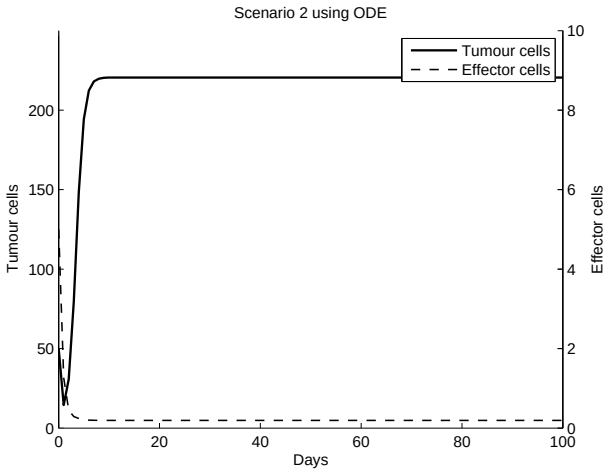


(b) SD

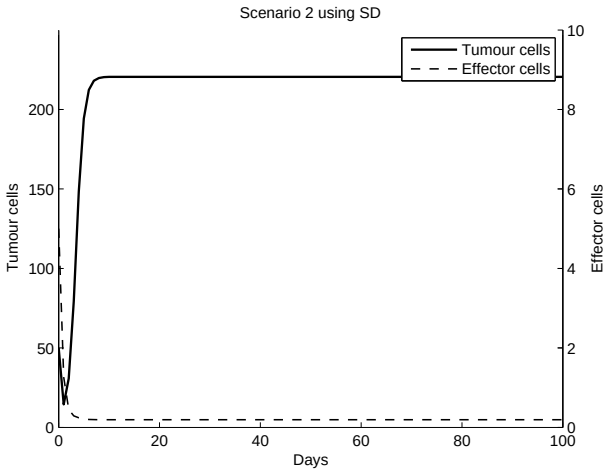


(c) ABMS

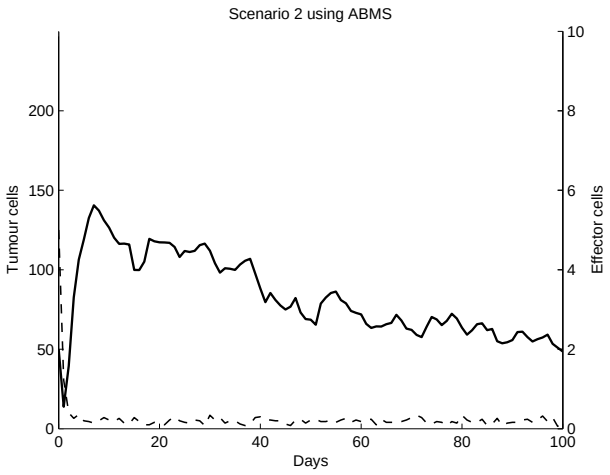
Figure 5.7: Results for scenario 1



(a) ODE

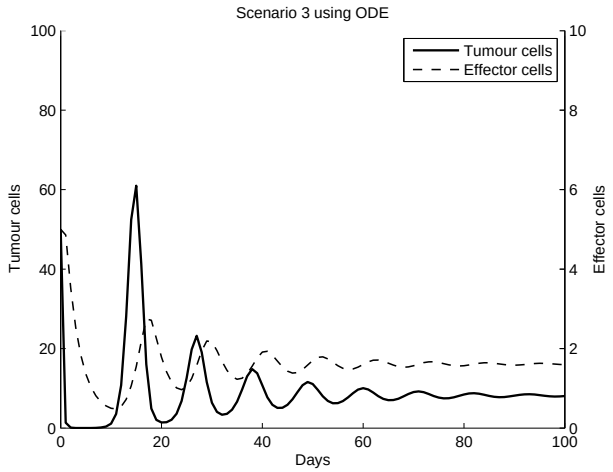


(b) SD

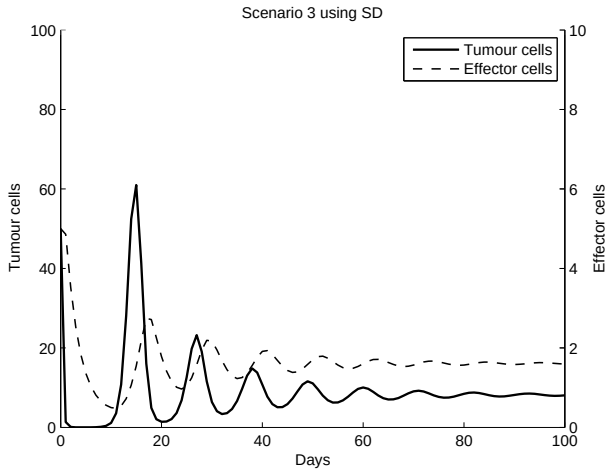


(c) ABMS

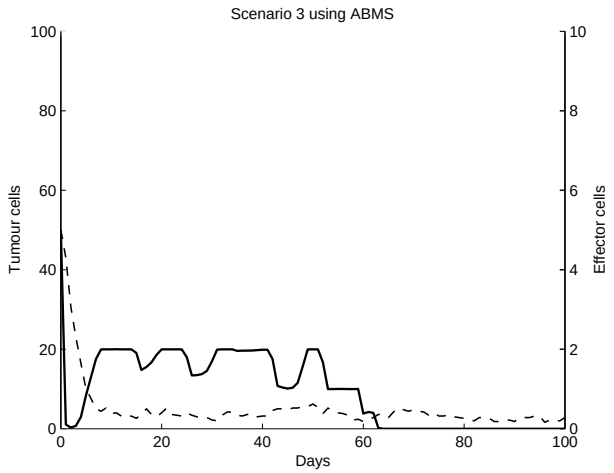
Figure 5.8: Results for scenario 2



(a) ODE

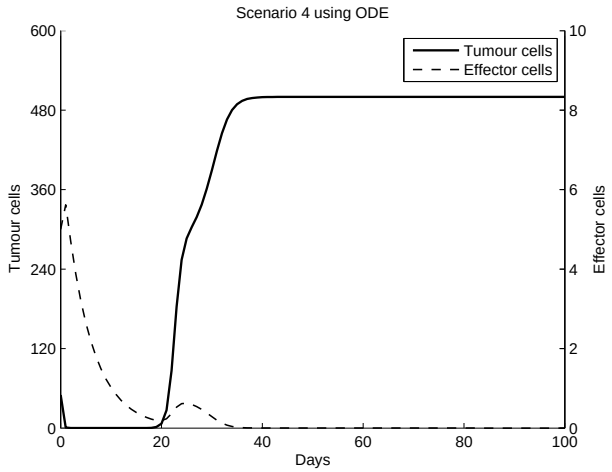


(b) SD

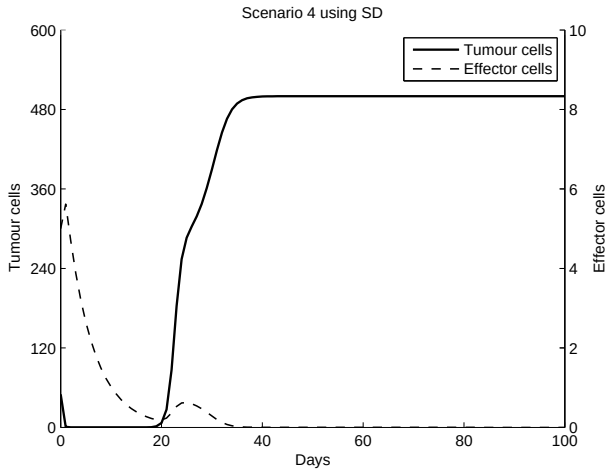


(c) ABMS

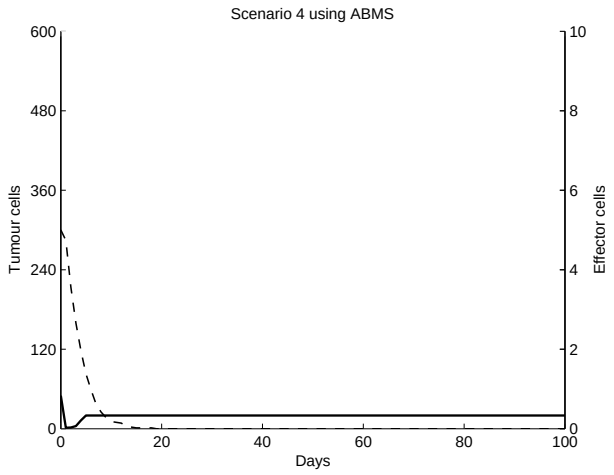
Figure 5.9: Results for scenario 3



(a) ODE



(b) SD



(c) ABMS

Figure 5.10: Results for scenario 4

5.2.6 Summary of Case 4 – Converting from System Dynamics to Agent-Based Modelling and Simulation

A mathematical model of tumour cells growth and their interactions with general immune effector cells was considered for conversion into SD and ABMS. Four experiments were conducted and, for only one of them, the results were similar to the mathematical model. The differences in the output were due to the fact that effector cells numbers changed continuously in the SD, while for the ABMS they changed in a discrete pattern.

The results in these experiments demonstrated that there were simulation cases where SD and ABMS derived from the same mathematical model do not have the same output. Without experimental data for validation, it is therefore impossible to conclude which approach would be more suitable for these cases.

One alternative would be the development of an ABMS solution, which is not based on the rates defined in the mathematical model. However, it would appear that, for each output (or parameter change on the mathematical model), there should be a different ABMS implementation. The constraint was therefore added that tumour cells should always be greater than zero in the ABMS for the second and fourth scenarios. The outputs became closer to those from the SD, as shown in Figure 5.11. This restriction, however, also changed the first scenario results which had previously seemed satisfactory. In scenario 4, although the outcome with this fix did not look similar at the start of the simulation, the steady state presented closer values.

In order to achieve closer results for scenario 3, the constraint was implemented that the number of effector cells should be greater than zero. The results are shown in line four of Figure 5.11. However, the fix did not work perfectly as only the steady state of the simulation presented similar results. Table 5.5 (page 150) shows the Wilcoxon statistical test results for fix 1 and fix 2.

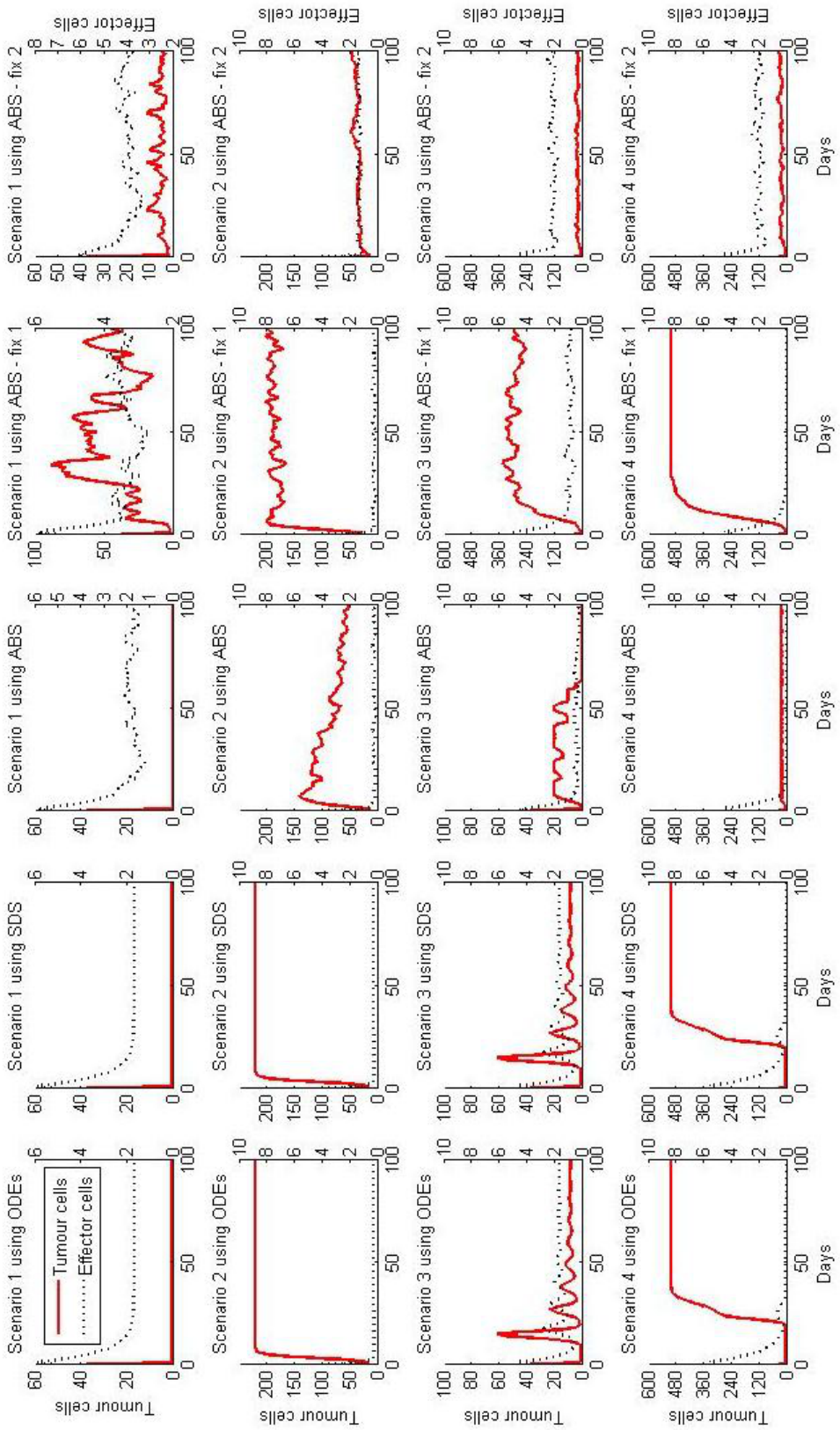


Figure 5.11: Results for case 4. In the first column the ODE results for the four scenarios are shown. The second column presents the SD results and the third column presents the ABMS results. The fourth and fifth columns show the fixes attempted to achieve similar outcomes. Fix 1 adds the constraint $tumourcells > 0$. Fix 2 adds the constraint $tumourcells > 0$ and $effectorcells > 0$. Each graph illustrates the results for tumour cells (continuous line) and effector cells (dotted lines). The scale for the results for tumour cells is on the left y axis of each graph while the scale for effector cells is on the right y axis.

5.2.7 From Agent-based Modelling and Simulation to System Dynamics

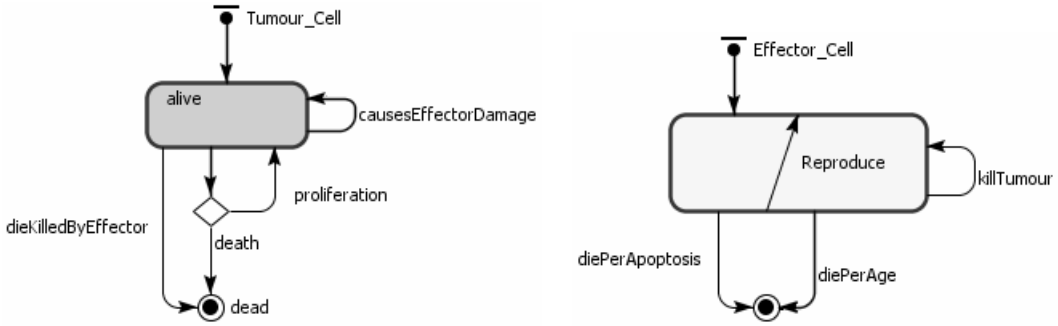
In this section we test our guidelines, as defined in Section 3.5.4, to convert the case 4 ABMS model into an SD model. We decided to use the same case study to convert from ABMS to SD in order to proceed the validation of the outcomes based on those from the mathematical model. Furthermore, this decision was based on the fact that we did not have access to other established ABMS models or experimental data to build an ABMS from scratch and then convert it to SD.

We performed our conversion from ABMS to SD as follows:

1. Identify the system structure. First there is the need to recognize the system structure and assume a high level of aggregation for the objects being modelled. It is necessary to generalise from the specific events and consider patterns of behaviour in the system. The cells, therefore, no longer respond individually. The simulation out- come is given by the collection of cells and its dynamics as a group. In our case, there are two cell populations (aggregations). The ABMS diagram (Figure 5.12) illustrates that the tumour cell population changes with time by proliferation, natural death and death caused by effector cells. The second population comprises the effector cells, which die with age or apoptosis/damage and reproduce. The effector cell population negatively impacts on the amount of tumour cells because effector cells kill tumour cells over time. The repro- duction of effector cells increases as the number of tumour cells increase. In addition, as effector cells kill tumour cells they become damaged. The tumour cell population therefore impacts on the effector cells population in both positive and negative ways.

2. Identify the stocks in the system. Stocks are physical entities which can accumulate over time. In our example, we defined as stocks the effector cells and tumour cells.

3. Define the stocks, their flows and information. Having the stocks (step



(a) Tumour cell agent (b) Effector cell agent

Figure 5.12: ABMS state charts for conversion to SD

2) and the knowledge about the structure of the model (step 1) we can depict how each stock is changed over time by the flows and the information about how a stock would influence a flow. The effector cells stock is subtracted by death and apoptosis. Moreover, it is increased by proliferation and treatment. Both death and proliferation require information about the stock of effector cells. The stock and flow diagram for effector cells is shown in Figure 5.13 below. The same occurs with the tumour cell stock, which is changed by proliferation and death.

4. Define the final stock and flow diagram. After defining the diagrams for each stock, it is necessary to return to the system structure and define how the stocks will interact or influence each other. As stated previously (step 1), tumour cells impact on the proliferation and death of effector cells, and effector cells influence the growth of tumour cells, as shown in Figure 5.13.

5. Define the mathematical model. For SD, a set of mathematical equations is necessary to describe how the stocks will change over time. The diagram depicted in Figure 5.13 indicates that the interactions between tumour cells and immune effector cells can be defined by the equations:

$$\frac{dT}{dt} = p_T(T) - d1_T(T) - d2_T(T, E) \tag{5.9}$$

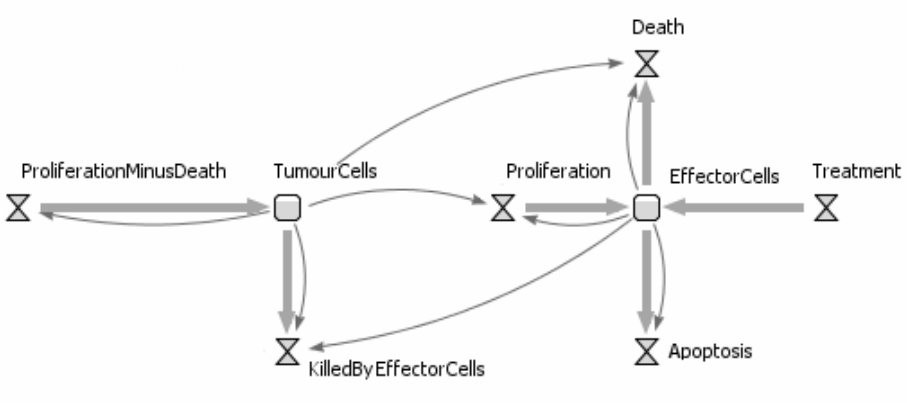


Figure 5.13: SD stock and flow diagram

$$\frac{dE}{dt} = p_E(T,E) - d_E(T,E) - a_E(E) \tag{5.10}$$

where T represents the number of tumour cells, E is the number of effector cells, $p_T(T) - d1_T(T)$ is the growth of tumour cells ($proliferation - death$), $d2_T(T,E)$ is the number of tumour cells killed by effector cells, $p_E(T,E)$ is the proliferation of effector cells, which is influenced by the number of tumour cells, and $a_E(E)$ is the death (apoptosis) of effector cells.

6. Define the parameters of the mathematical model. The parameters are obtained from the transition rates (Table 5.6):

where $a = 1.636$, $g = 20.19$, $m = 0.00311$, $n = 1$ and $p = 1.131$. The parameters b, d and s vary according to the scenario studied.

7. Define the flow calculations A flow expression is defined by looking at the transition rate calculation and the information defined in the stock and flow diagram, as illustrated in Table 5.7 below. The index i in the formulas indicates that the variable is obtained by referring to the information in the stock and flow diagram of Figure 5.13.

8. Define the final SD model. The final SD model, containing the stocks, flows, information and parameters is shown in Figure 5.14. The resulting stock and flow diagram is the same as that from Figure 5.5, which verifies the effectiveness of our

Table 5.6: Transition rates from ABMS model for case 4

Agent	Transition	Transition rate	Parameter(s)
Tumour Cell	Proliferation	$a - (TotalTumourCells.b)$	a and b
	DieWithAge	$a - (TotalTumour.b)$	a and b
	DieKilledByEffectorCells	message from effector	n
Effector Cell	Proliferation	$\frac{p.TotalTumourCells}{g+TotalTumourCells}$	p and g
	Die	d	d
	DieWithAge	$m.TotalTumourCells$	m
	KillTumour	n	n

Table 5.7: Flow calculations for case 4 from ABMS to SD

Stock	Flow	Flow formula
TumourCells	$ProliferationMinusDeath$	$TumourCells_i(a(1 - b.TumourCells))$
	$KilledByEffectorCells$	$n.TumourCells_i.EffectorCells$
EffectorCells	$Proliferation$	$\frac{p.TumourCells.EffectorCells}{g+TumourCells}$
	$Death$	$m.TumourCells.EffectorCells_i$
	$Apoptosis$	$d.EffectorCells_i$
	$Treatment$	s

guidelines for this case.

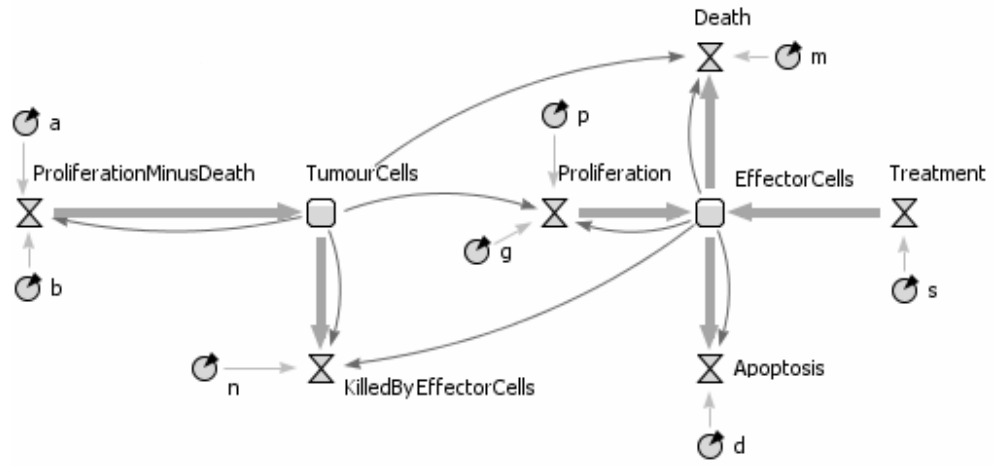


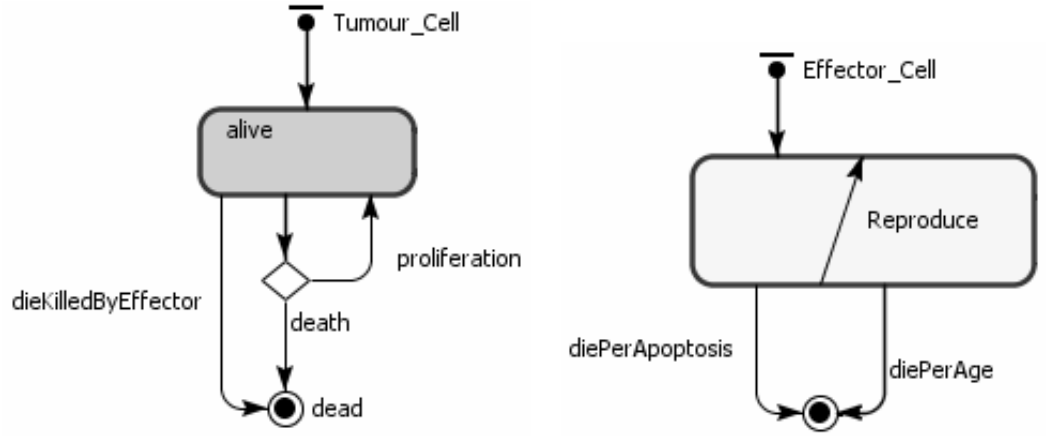
Figure 5.14: SD model

5.2.8 From Agent-based Modelling and Simulation to System Dynamics Considering Agents' Movement

In the previous section the model considered is non-spatial and therefore the effector cells do not move to reach a tumour cell. The non-spatial character of the model impacts on the results of the simulations because tumour cells die at a rate that is calculated based on the entire population of effector cells. In order to construct a simulation of a system closer to reality, a certain effector cell E_{c_i} has to move towards a tumour cell T_{c_i} and kill it. The remaining effector cells in the population will therefore have no impact on the death of T_{c_i}. There is now a new simulation, in which cellular spatial location is considered. The objectives with this new scenario are: (1) to observe the differences in the model's behaviour over time, compared with the previous static model; and (2) to investigate how the movement of effector cells would impact on the guidelines for the SD model construction.

The Agent-based Model

The agents' state charts of the dynamic model are similar to those considered in the static model, as shown in Figure 5.15:



(a) Tumour cell agent (b) Effector cell agent

Figure 5.15: ABMS state charts for case 4 considering agent's movement

Concerning the agents' behaviours, those from the static model (Table 5.2) remain. In addition, the random movement behaviour is considered for both effector cell and tumour cell agents. The movement of the agents is controlled by events that occur at a certain rate (in our simulations, 0.01 was the rate for effector cell movement and 0.0001 for tumour cell movement). At each step of the simulation, an effector cell agent will look for a tumour cell and move towards it in order to kill it. In order to do the killing, an effector cell will send a message to the corresponding tumour cell agent, which will then die. The remaining transitions are triggered by the corresponding rates.

System Dynamics Model for the Dynamic cells of the Agent-based Model

At each time step in the simulation, the maximum number of tumour cells killed is equal to the number of effector cells, given the movement constraint added. Equation 5.9 is therefore replaced by Equation 5.11:

$$\frac{dT}{dt} = p_T(T) - d1_T(T) - d2_T(E) \tag{5.11}$$

where $d2_T(E) = nE$

The final SD model for the dynamic agent-based model will differ from that for the static agent-based model with regard to the $KilledByEffectorCells$ flow, which will no longer have the information from the tumour cells stock, as shown in Figure 5.16:

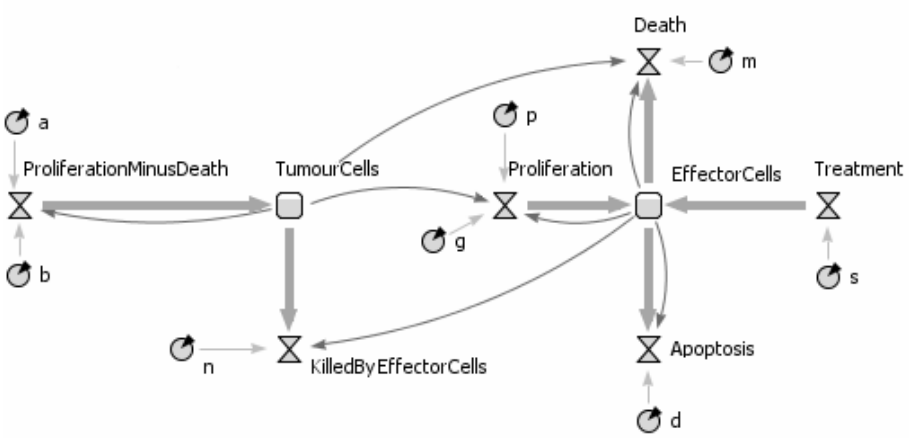


Figure 5.16: SD model corresponding to the dynamic agent-based model

5.2.9 Experiments

Three scenarios were considered in our experiments, as depicted in Table 5.8 below. These were similar scenarios to those for the previous experiments (Section 5.2.4); however, treatment was not considered.

Scenario	b	d
1	0.002	0.1908
2	0.004	2
3	0.002	0.3743

Table 5.8: Simulation parameters for different scenarios in the dynamic models. For the other parameters, the values are the same in all experiments, i.e. $a = 1.636$, $g = 20.19$, $m = 0.00311$, $n = 1$ and $p = 1.131$.

The simulations were run for a period equivalent to one hundred days, using both approaches. Fifty replications for the ABMS were run and the mean values for the outputs were collected.

5.2.10 Results for the Static Model

The results obtained for the static model are the same as those from Figure 5.2.6, as the SD obtained from the ABMS is the same as that obtained from the mathematical model.

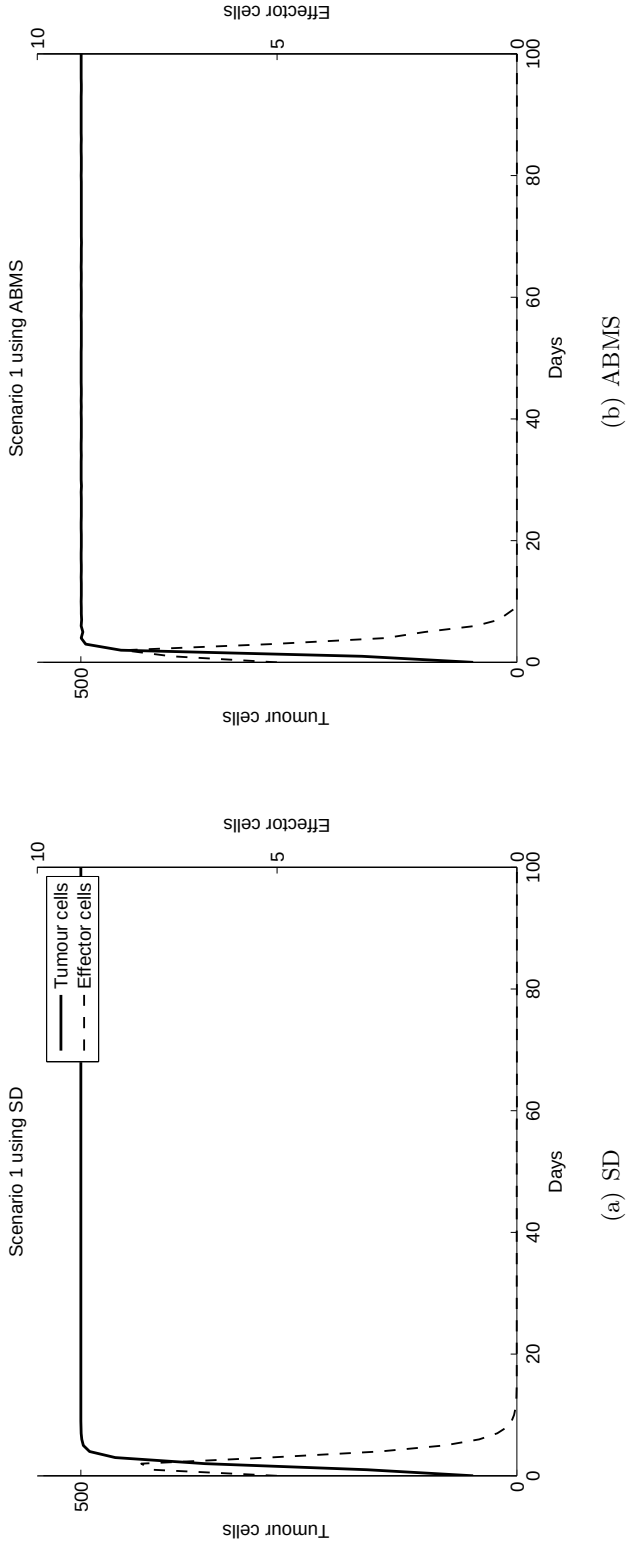
5.2.11 Results for the Dynamic Model

The simulation results are shown in Figures 5.17, 5.18 and 5.19. For all scenarios, the results for tumour cells are very similar in both approaches. In the third scenario, differences occur in the initial growth of effector cells, which makes the maximum number of this population greater than the SD. The steady-state is very similar for both approaches.

5.2.12 Summary of Case 4 – Converting from Agent-Based Modelling and Simulation to System Dynamics

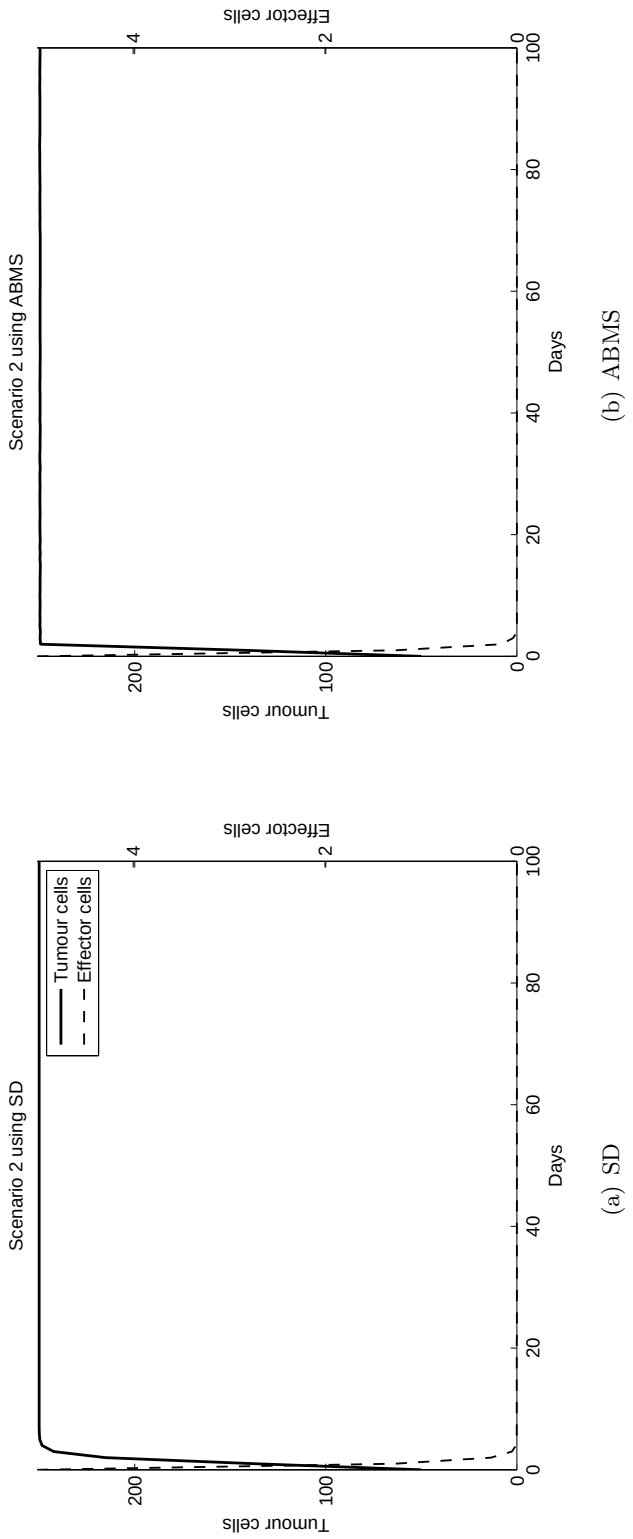
In the previous sections we tested our guidelines in order to convert from ABMS models to SD models. Case 4 ABMS model obtained from the SD was reverted back to SD. We decided to use the same case study to convert from ABMS to SD in order to proceed the validation of the outcomes based on those from the mathematical model. Furthermore, we did not have access to other established ABMS models or experimental data to build an ABMS from scratch and then convert it to SD.

In addition to the original non-spatial model, we defined a hypothetical system that considered the spatial movement of agents. Results indicated that it is possible to obtain a SD model based on the information inherited in an ABMS model for our case



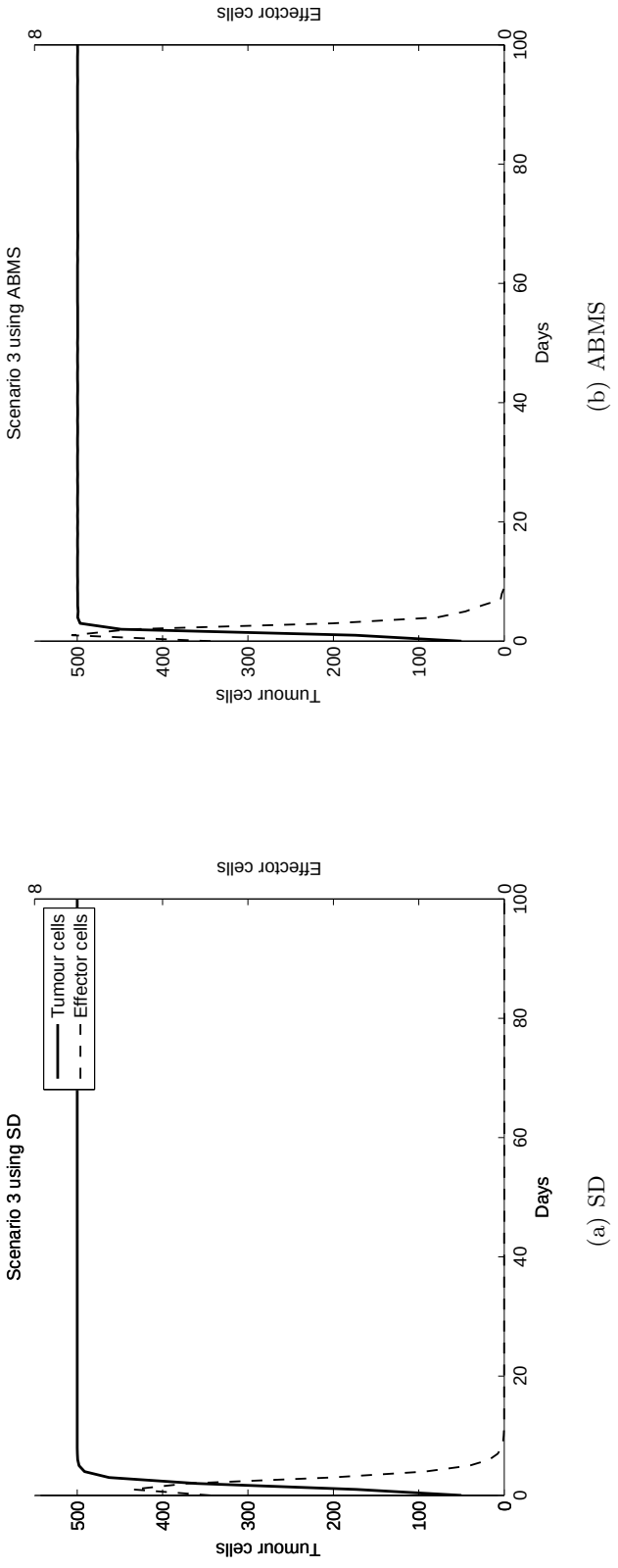
(a) SD (b) ABMS

Figure 5.17: Results for scenario 1 considering cellular movement



(a) SD (b) ABMS

Figure 5.18: Results for scenario 2 considering cellular movement



(a) SD (b) ABMS

Figure 5.19: Results for scenario 3 considering cellular movement

study, however, the outcomes are not always similar.

For the non-spatial scenario, results differed because stock numbers changed continuously in the SD, while for the ABMS agents' quantities changed in a discrete pattern. Furthermore, the variability produced by in the outcomes from the ABMS was not replicable in the obtained SD model. These results are important as they exemplify circumstances in which differences in the outcomes should be expected and why they occur. In addition, it is necessary to evaluate together with experts in immunology whether these differences are acceptable when compared to their real-world experimentation. This outcome information should also be considered when there is the necessity of re-conceptualizing a model in a multi-paradigm approach, given the impact of translating between paradigms.

In the spatial scenario, tumour cell outcomes were very similar for both approaches; however, the effector cell results for one scenario were slightly different as the numbers in the ABMS were greater than the SD values. Such differences are due to the randomness and discrete values for agents in the ABMS. This experiment showed that there are examples where it is possible to represent movement with SD, which is useful for multi-paradigm modelling.

5.3 Case 5: Interactions Between Tumour Cells, Effector Cells and Cytokines IL-2

The second case study investigated in this chapter is concerned with a mathematical model for the interactions between tumour cells, effector cells and the cytokine IL-2. This is an extension of the previous study since it considers IL-2 as molecules that will mediate the immune response towards tumour cells. They will affect the proliferation of effector cells according to the number of tumour cells in the system. Treatment is now applied in two different ways, by injecting effector cells or injecting cytokines. The details of the mathematical model are introduced in the following section.

5.3.1 The Mathematical Model

The mathematical model we use in case 5 is obtained from [42]. The model's equations illustrate the non-spatial dynamics between effector cells (E), tumour cells (T) and the cytokine IL-2 (I_L), described by the following differential equations:

$$\frac{dE}{dt} = cT - \mu_2 E + \frac{p_1 E I_L}{g_1 + I_L} + s1 \tag{5.12}$$

Equation 5.12 describes the rate of change for the effector cell population E [42]. Effector cells grow based on recruitment (cT) and proliferation ($\frac{p_1 E I_L}{g_1 + I_L}$). The parameter c represents the antigenicity of the tumour cells (T) [1, 42]. μ_2 is the death rate of the effector cells. p_1 and g_1 are parameters used to calibrate the recruitment of effector cells and $s1$ is the treatment that will boost the number of effector cells.

$$\frac{dT}{dt} = a(1 - bT) - \frac{a_a E T}{g_2 + T} \tag{5.13}$$

Equation 5.13 describes the changes that occur in the tumour cell population T over time. The term $a(1 - bT)$ represents the logistic growth of T (a and b are parameters that define how the tumour cells will grow) and $\frac{a_a E T}{g_2 + T}$ is the number of tumour cells

killed by effector cells. a_a and g_2 are parameters to adjust the model.

$$\frac{dI_L}{dt} = \frac{p_2 ET}{g_3 + T} - \mu_3 I_L + s2 \tag{5.14}$$

The IL-2 population dynamics is described by Equation 5.14. $\frac{p_2 ET}{g_3+T}$ determines IL-2 production, which is regulated by the parameters p_2 and g_3 [42]. μ_3 is the IL-2 loss. $s2$ also represents treatment, which is given by the injection of IL-2 in the system.

5.3.2 The System Dynamics Model

From Ordinary Differential Equations to System Dynamics

Model Stocks

The SD model contains three stock variables: tumour cells, effector cells and IL-2.

Model Flows and Information

The stock of effector cells, described by Equation 5.12, is changed by the recruitment of new effector cells, according to the number of tumour cells, death, proliferation and treatment (insertion of new effector cells). There is information from the effector cells stock to proliferation and death flows, as shown in Figure 5.20:

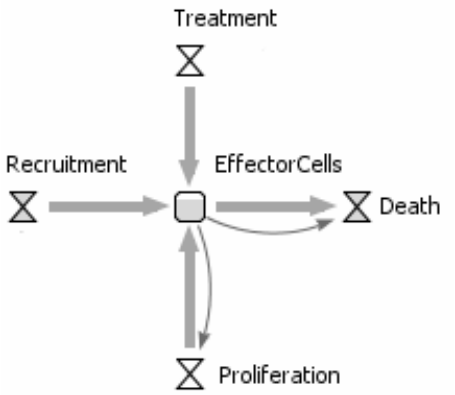


Figure 5.20: The effector cell stock variable with its flows and information

Equal to the tumour cell stock of case 4, this is changed by its natural proliferation and death as well as by the number of cells killed by effector cells (Figure 5.3).

IL-2 stock changes with the production of new IL-2 molecules from effector cells (the production also depends on the number of tumour cells), loss and treatment (insertion of IL-2). The production of IL-2 depends on the number of tumour cells, and the loss of IL-2 is proportional to its quantity in the system, as shown in Figure 5.21:

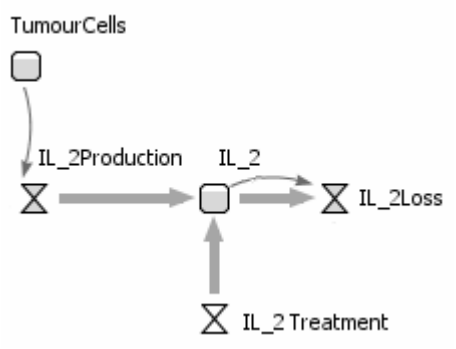


Figure 5.21: The IL-2 stock variable with its flows and information

Model Parameters

The model parameters are the same as those from the mathematical model, i.e. c, μ_2, p_1, g_1, $s1$, a, b, a_a, g_2, p_2, g_3, μ_3 and s_2.

Flows Calculations

Table 5.9 below shows how the equations from the mathematical models are defined for the flow values.

The Final System Dynamics Model

The final SD stock and flow diagram is depicted in Figure 5.22 below.

Table 5.9: Flow calculations for case 5

Stock	Flow	Expression	Flow formula
EffectorCells	$Proliferation$	$\frac{p_1 EI_L}{g1+I_L}$	$\frac{p_1.IL\_2.EffectorCells}{g1+IL\_2}$
	$Death$	$\mu_2 E$	$mu2 \times EffectorCells$
	$Recruitment$	cT	$c \times TumourCells$
	$Treatment$	s_1	$s1$
TumourCells	$ProliferationMinusDeath$	$a(1-bT)T$	$TumourCells \times (a(1-b.TumourCells))$
	$KilledByEffectorCells$	$\frac{a_a ET}{g_2+T}$	$\frac{aa.TumourCells.EffectorCells}{g2+TumourCells}$
IL_2	$IL\_2Production$	$\frac{p_2 ET}{g_3+T}$	$\frac{p2.EffectorCells.TumourCells}{g3+TumourCells}$
	IL_2Loss	$\mu_3 I_L$	$mu3 \times IL\_2$
	$IL\_2Treatment$	s_2	$s2$

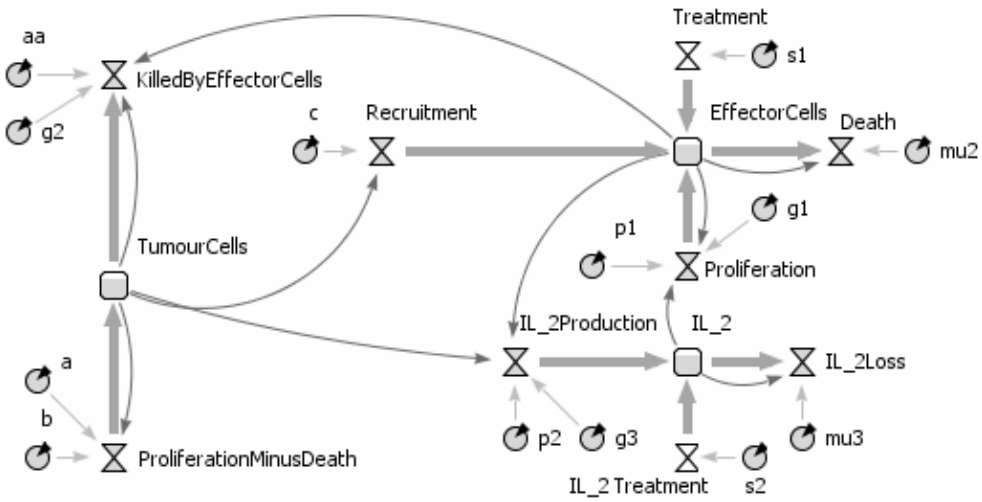


Figure 5.22: SD model for case 5

5.3.3 From System Dynamics to Agent-based Modelling and Simulation

Model Agents

Our agents correspond to the stocks in the SD model. The populations of agents are therefore the effector cells, tumour cells and IL-2.

Agents' Behaviours

The behaviour of each agent is shown in Table 5.10.

Table 5.10: Agents' parameters and behaviours for case 5

Agent	Parameters	Reactive behaviour	Proactive behaviour
Effector Cell	$mu2$	Dies	
	$p1$ and $g1$		Reproduces
	c	Is recruited	
	$s1$	Is injected as treatment	
	$p2$ and $g3$		Produces IL-2
	aa and $g2$		Kills tumour cells
Tumour Cell	a and b	Dies	
	a and b		Proliferates
	aa and $g2$	Dies killed by effector cells	
	c		Induces effector recruitment
IL-2	$p2$ and $g3$	Is produced	
	$mu3$	Is lost	
	$s2$	Is injected	

Agent Implementation

Based on the agents, parameters and behaviours derived from the previous step, state charts for each agent type were developed, as shown in Figure 5.23.

The ABMS model rates corresponding to the flow values in the SD model are given in Table 5.11 below. In the transition rate calculations, the variable $TotalTumour$

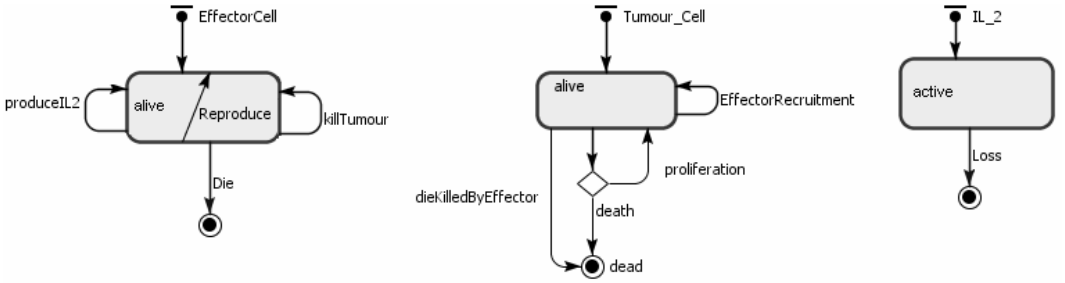


Figure 5.23: ABS state charts for the agents of case 5

corresponds to the total number of tumour cell agents, the variable $TotalEffector$ is the total number of effector cell agents and $TotalIL\_2$ is the total number of IL-2 agents.

Table 5.11: Transition rates calculations from SD flows equations for case 5

Agent	Transition	SD Flow equation	Transition rate
Effector Cell	Reproduce	$\frac{p_1.IL\_2.EffectorCells}{g1+IL\_2}$	$\frac{p_1.TotalIL\_2.TotalEffector}{g1+TotalIL\_2}$
	Die	$EffectorCells.mu2$	$mu2$
	killTumour	$\frac{aa.TotalEffector.TotalTumour}{g2+TotalTumour}$	$aa\frac{TotalTumour}{g2+TotalTumour}$
	ProduceIL2	$\frac{p2.EffectorCells.TumourCells}{g3+TumourCells}$	$\frac{p2.TotalTumour}{g3+TotalTumour}$
umour Cell T	Reproduce	$a.TumourCells(1-TumourCells.b)$	$a-(TotalTumour.b)$
	Die	$a.TumourCells(1-TumourCells.b)$	$a-(TotalTumour.b)$
	DieKilledByEffector	$\frac{aa.TumourCells.EffectorCells}{g2+TumourCells}$	message from effector
IL-2	Loss	$IL\_2.mu3$	$mu3$

Simulation

For the simulation building, apart from the agents, there are also two events:

1. $TreatmentS1$, which adds effector cell agents according to the parameter $s1$

2. $TreatmentS2$, which adds IL-2 agents according to the parameter $s2$

5.3.4 Experiment

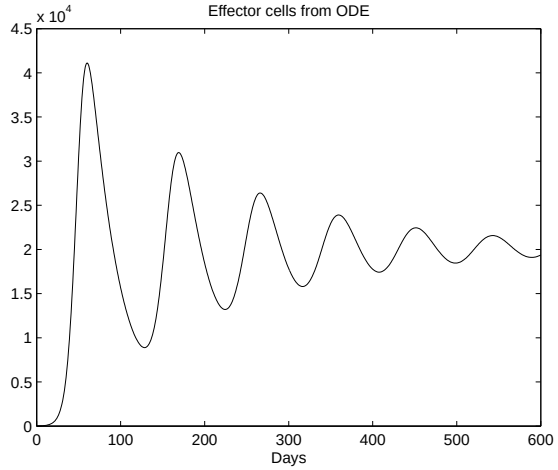
The experiment was conducted assuming the parameters of Table 5.12:

Table 5.12: Parameter values for case 5

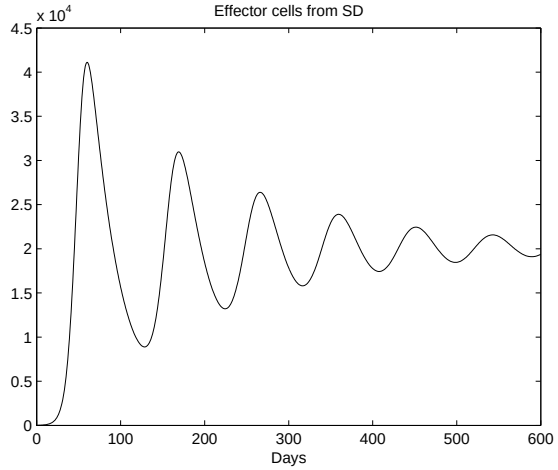
Parameter	Value
a	0.18
b	0.000000001
c	0.05
aa	1
g2	100000
s1	0
s2	0
mu2	0.03
p1	0.1245
g1	20000000
p2	5
g3	1000
mu3	10

5.3.5 Results

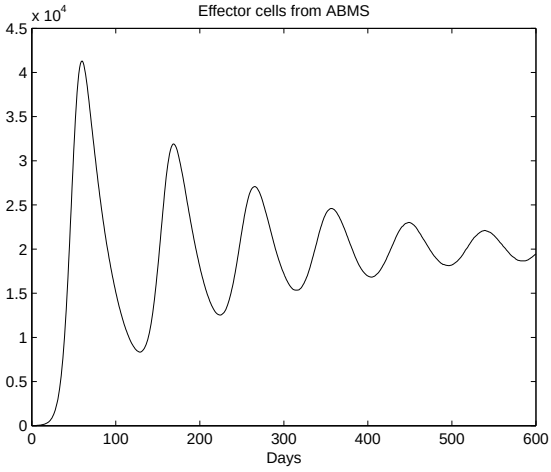
The results obtained are shown in Figures 5.24, 5.25 and 5.26 for effector cells, tumour cells and IL-2 respectively. The SD and ABMS were validated by comparing its outputs with those produced by the ODEs. As the figures reveal, the results for all populations are very similar; the growth and decrease of all populations occur at similar times for both approaches. Furthermore because of the large population sizes (around 10^4), ABMS model curves have minor erratic behaviour, which corroborates to the similar patterns observed in the outcomes. These similarities are also confirmed by the Wilcoxon test results presented in Table 5.13. The table shows the p-values obtained with a 5% significance level. For the effector and tumour cells, the p-value was higher than 0.5, which indicates that the test failed to reject the null hypothesis that the outcomes were similar. As the overall results are very close, in this case the use of SD is preferable.



(a) ODE

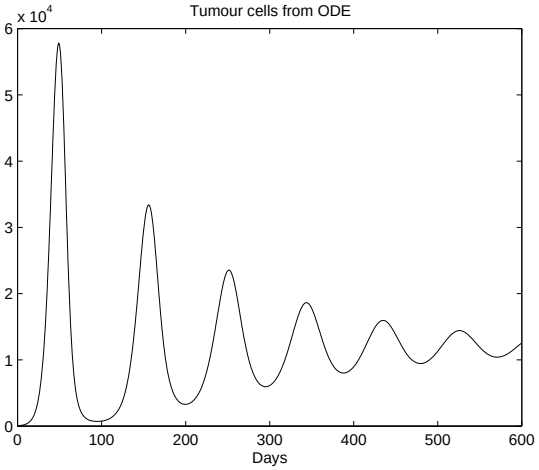


(b) SD

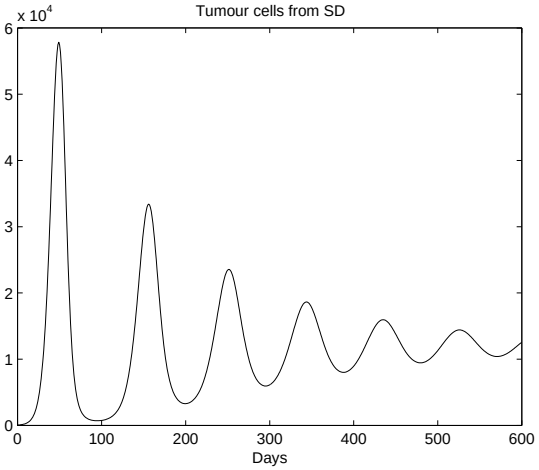


(c) ABMS

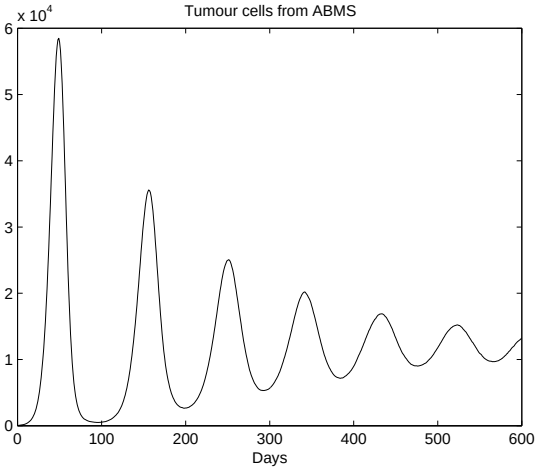
Figure 5.24: SD and ABMS results for effector cells



(a) ODE

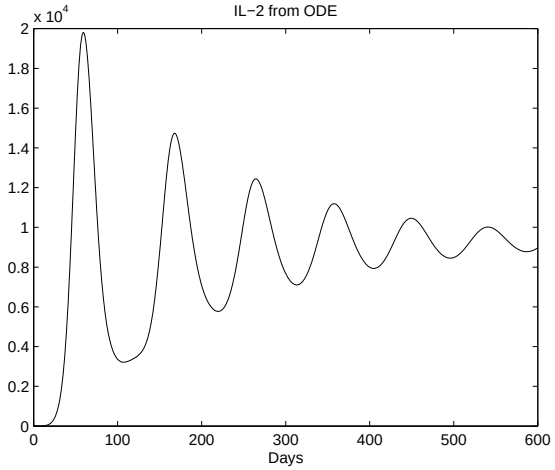


(b) SD

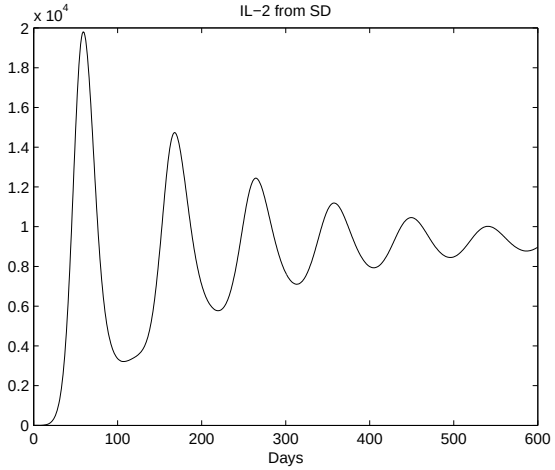


(c) ABMS

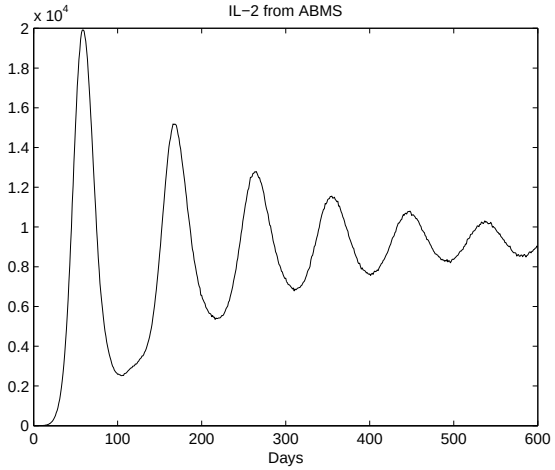
Figure 5.25: SD and ABMS results for tumour cells



(a) ODE



(b) SD



(c) ABMS

Figure 5.26: SD and ABMS results for IL2

Table 5.13: Wilcoxon test with 5% significance level comparing the results from SD and ABMS for case 5

Population	p
Effector	0.7231
Tumour	0.5710
IL2	0.4711

5.3.6 From Agent-based Modelling and Simulation to System Dynamics

In this section we test our guidelines, as defined in Section 3.5.4, to convert an ABMS model into an SD model. As this has been achieved in the previous case study, an ABMS including cellular and molecular movement is considered. The results obtained from experimentation considering the conversion from ABMS to SD for static model are the same as those from the previous section, as the SD obtained from the ABMS is the same as that obtained from the mathematical model. In this section, therefore, we focus on the dynamic model.

In order to construct a simulation of a system closer to reality, a certain effector cell E_{c_i} has to move towards a tumour cell T_{c_i} and kill it. The remaining effector cells in the population therefore will not have any impact on the death of T_{c_i}. Furthermore, an effector cell E_{c_j} only proliferates when it gets in touch with an IL-2 molecule, which moves towards it.

The Agent-based Model

The agents' state charts of the dynamic model are the same as those considered in the static model (Figure 5.23 of Section 5.3.3). Regarding the agents' behaviours, those from the static model (Table 5.10) remain. In addition, the random movement behaviour is considered for effector cell, tumour cell and IL-2 agents. The movement of the agents is controlled by events that occur at a certain rate (in our simulations, 0.01 was the rate for effector cell and IL-2 movement and 0.0001 for tumour cell movement). At each step of the simulation, an effector cell agent will seek for a tumour cell and

move towards it in order to kill it. In addition, an IL-2 will move towards an effector cell to allow for its replication. In order to do the killing, an effector cell will send a message to the corresponding tumour cell agent, which will then die. The transition $dieKilledByEffectorCells$ will now therefore be triggered by this message. In addition, the proliferation rate of the effector cells will be greater than zero only when this cell meets the IL-2 (distance between them equal to zero). The remainder transitions are still triggered by the corresponding rates, defined in the static agent-based model. As new requirements were added to the model, there is no data or mathematical model to validate it. Our goal therefore is to verify if we still can obtain an equivalent SD model and test our guidelines.

System Dynamics Model

At each time step in the simulation, the maximum number of tumour cells killed is equal to the number of effector cells, given the movement constraint added. The tumour cell dynamics is therefore described by Equation 5.15:

$$\frac{dT}{dt} = p_T(T) - d1_T(T) - d2_T(E) \tag{5.15}$$

where: $d2_T(E) = aaE$

Effector cells only proliferate if they meet an IL-2 molecule. In this case, therefore, there will be two types of effector cells: those in proliferation and those that do not proliferate. Hence, the effector cells equation should be:

$$\frac{dE}{dt} = E_{np} + E_p - \mu_2 E \tag{5.16}$$

where:

$$E_{np} = cT + s1 + \frac{p_1 E_p I_L}{g_1 + I_L} \tag{5.17}$$

$$E_p = I_L E_{np} \tag{5.18}$$

The final SD model for the dynamic agent-based model is shown in Figure 5.27. As IL-2 molecules disappear from the system once they encounter an effector cell, the outflow *AbsorbedByEffector* is added to the IL-2 stock. We split the effector cell population in two stocks: the one that considers cells that are stimulated by IL-2 to reproduce *EffectorCellsInProliferation* and the stock which regards effector cells without proliferation *EffectorCellsNoProliferation*. The number of tumour cells is influenced by both populations of effector cells and the flow *TumourCellsKilledByEffectorCells* calculation which best suits our experiments is equal to the minimal element of

$$[TumourCells, (EffectorCellsInProliferation + EffectorCellsNoProliferation)].$$

Cells from the stock *EffectorCellsNoProliferation* migrate to the stock *EffectorCellsInProliferation* via flow $MeetIL_2$ according to the minimal element of

$$[EffectorCellsNoProliferation, IL - 2].$$

The division of the effector cells stock was not considered in our guidelines although it would appear to be a suitable approach for this case. It suggests that in situations where the agents change their behaviours, there should be a corresponding stock for each behaviour.

Experiment

The parameter values considered were the same as those from the static model. The simulations were run for a period equivalent to one hundred days using both approaches. Fifty replications for the ABMS were run and the mean values for the outputs were collected.

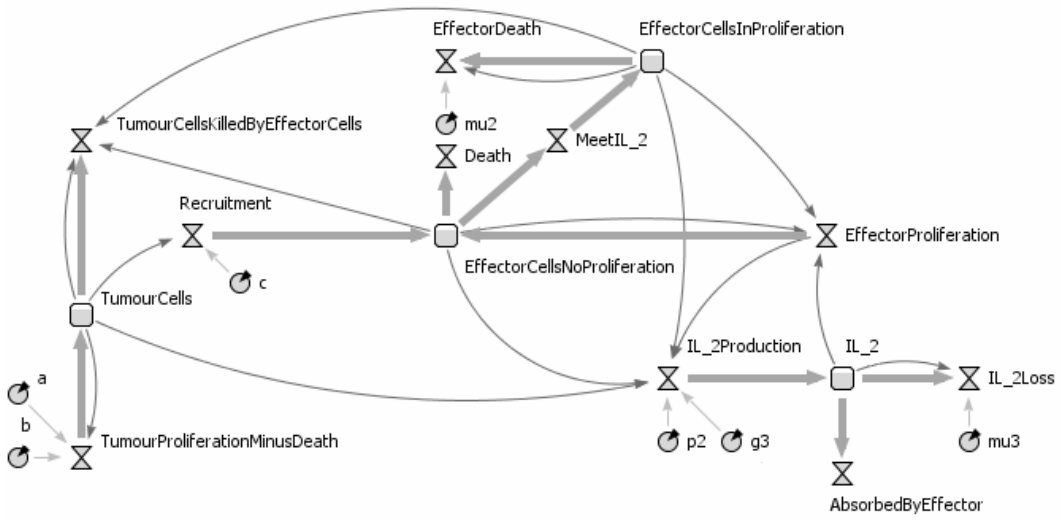
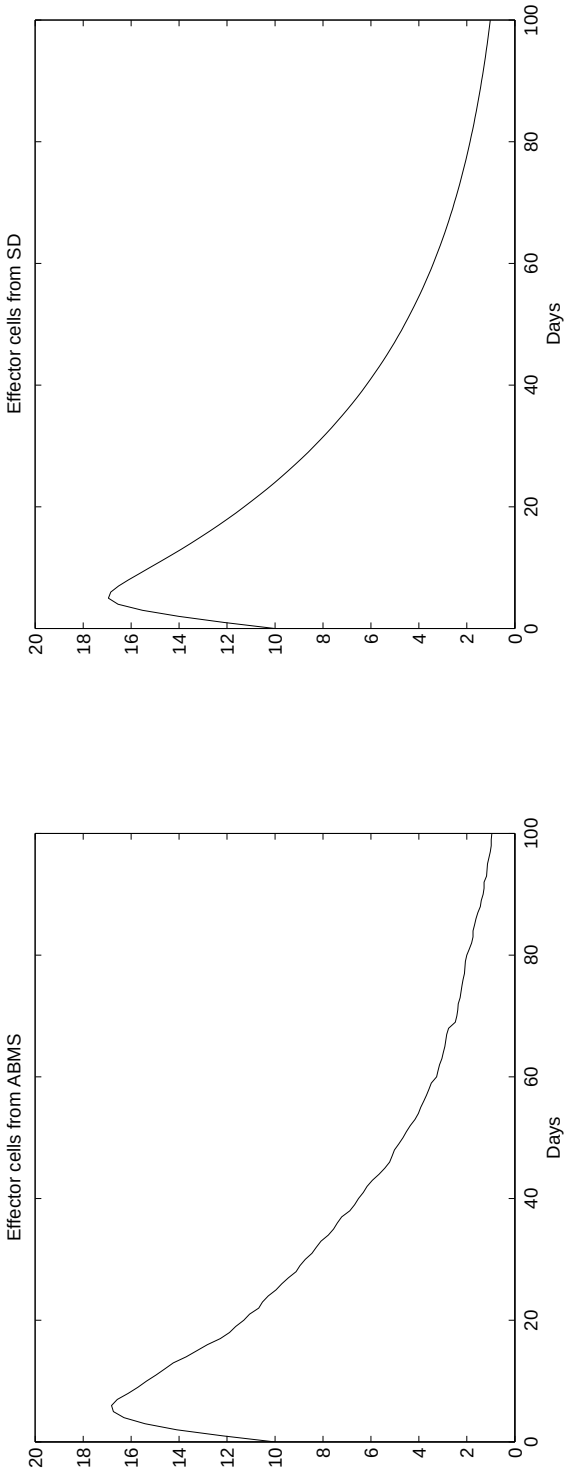


Figure 5.27: SD model corresponding to the dynamic agent-based model for case 5

Results

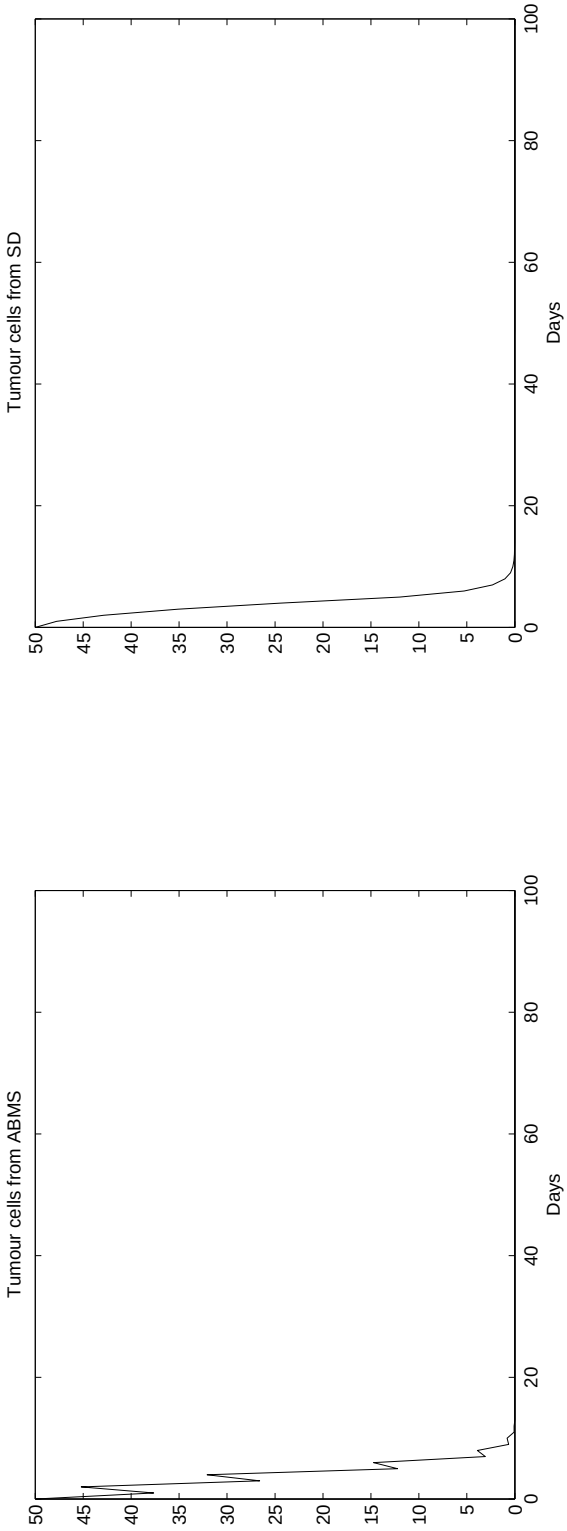
The simulation results are given in Figures 5.28 (for effector cells), 5.29 (for tumour cells) and 5.30 (for IL-2).

Regarding the effector cells outcomes, the results of both approaches are very close, confirmed by the p-value equal to 0.9493 obtained with the Wilcoxon test. For tumour cells and IL-2, however, the results differ due to the erratic behaviour of the ABMS outcome curves, which is explained by its stochasticity.



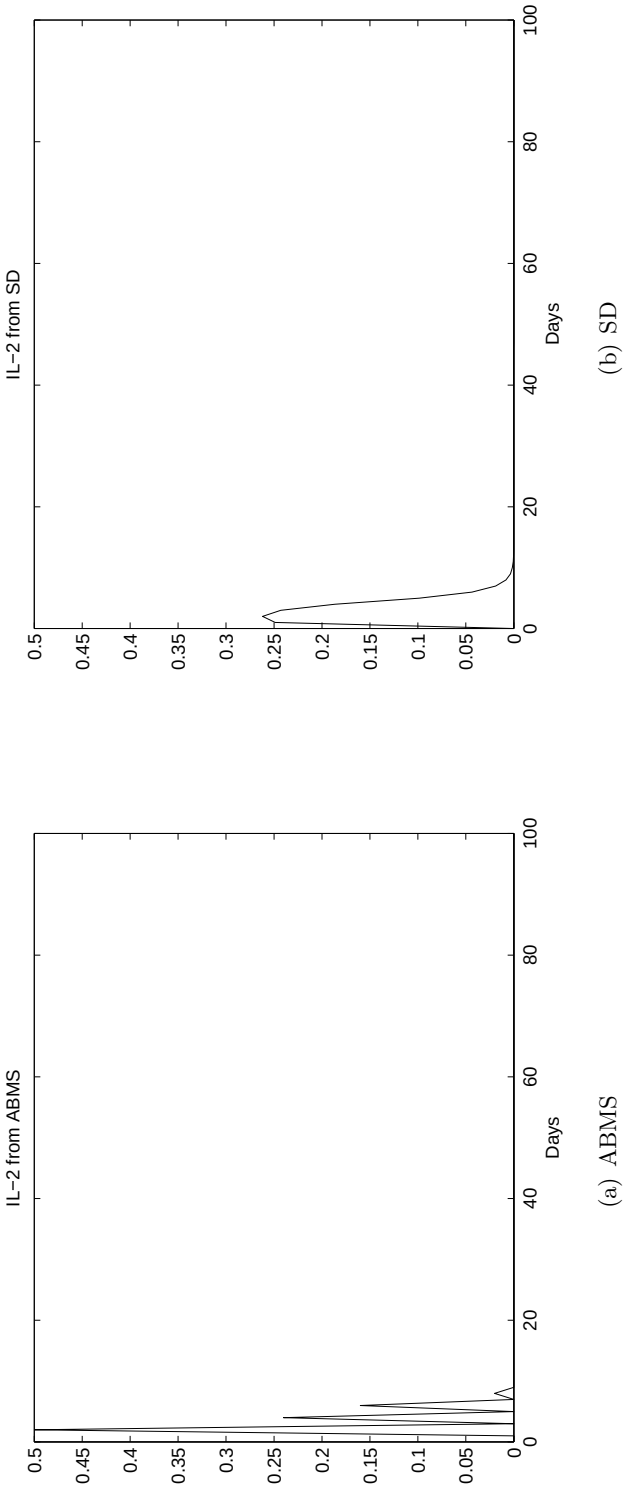
(a) ABMS (b) SD

Figure 5.28: Results for effector cells considering cellular movement



(a) ABMS (b) SD

Figure 5.29: Results for tumour cells considering cellular movement



(a) ABMS (b) SD

Figure 5.30: Results for IL-2 considering cellular movement

Summary of Case 5

Section 5.3.6 considered a scenario involving interactions between tumour cells, effector cells and cytokines IL-2. Furthermore the movement of these cells and molecules was implemented for the simulations. The objective was to build an ABMS simulation implementing the agents' movement and subsequently converting this model into an SD model. In addition, the effectiveness of the guidelines to convert between ABMS and SD was evaluated.

Regarding the conversion guidelines, a new scenario emerged that had not previously been considered: some of the agents' behaviours changed during the course of the simulation. Specifically for our case study, effector cells had an initial reproduction rate equal to zero. After they encountered an IL-2 molecule, their reproduction rate was set to a value greater than zero. Although these behaviours could have been implemented as different states of the same agent, only one state was deliberately defined and the corresponding rate after the cell molecule encounter occurred. If there were one state for effector cells reproducing and another state for effector cells not reproducing, the guidelines as defined would be enough for the conversion. By defining only one state, however, there was the need to review the guidelines to suit this new ABMS implementation. For this case, therefore, when agents change behaviour, it is necessary to define a stock variable for each new behaviour that occurs during the course of the simulation. The results of both approaches with the conversion are very similar, which indicates that the guidelines for this case study are effective. It is certainly necessary to acknowledge the variances that occur in the ABMS which are not replicated in the SD.

5.4 Case 6: Interactions Between Tumour Cells, Effector Cells, IL-2 and TGF-β

The third case study is based on the mathematical model of Arciero *et al.* [1], which consists of a system of ordinary differential equations describing interactions between tumour cells and immune effector cells, as well as the immune-stimulatory and suppressive cytokines IL-2 and TGF-β. According to Arciero *et al.* [1] TGF-β stimulates tumour growth and suppresses the immune system by inhibiting the activation of effector cells and reducing tumour antigen expression. The mathematical model, together with further details on the interactions studied is introduced in the following section.

5.4.1 The Mathematical Model

The mathematical model we use in case 6 is obtained from [42]. The model's equations illustrate the non-spatial dynamics between effector cells (E), tumour cells (T), IL-2 (I) and TGF-β (S) cytokines. The model is described by the following differential equations:

$$\frac{dE}{dt} = \frac{cT}{1+\gamma S} - \mu_1 E + \left(\frac{p_1 EI}{g_1 + I}\right)\left(p_1 - \frac{q_1 S}{q_2 + S}\right) \tag{5.19}$$

Equation 5.19 describes the rate of change for the effector cell population E. According to [1], *effector cells are assumed to be recruited to a tumour site as a direct result of the presence of tumour cells.* The parameter c in $\frac{cT}{1+\gamma S}$ represents the antigenicity of the tumour, which measures the ability of the immune system to recognize tumour cells. The presence of TGF-β (S) reduces antigen expression, thereby limiting the level of recruitment, measured by inhibitory parameter γ. The term $\mu_1 E$ represents loss of effector cells due to cell death, and the proliferation term $\left(\frac{p_1 EI}{g_1+I}\right)\left(p_1 - \frac{q_1 S}{q_2+S}\right)$ asserts that effector cell proliferation depends on the presence of the cytokine IL-2 and is decreased when the cytokine TGF-β is present. p_1 is the maximum rate of effector cell proliferation in the absence of TGF-β, g_1 and q_2 are half-saturation constants, and

q_1 is the maximum rate of anti-proliferative effect of TGF-β.

$$\frac{dT}{dt} = aT\left(1 - \frac{T}{K}\right) - \frac{a_a ET}{g_2 + T} + \frac{p_2 ST}{g_3 + S} \tag{5.20}$$

Equation 5.20 describes the dynamics of the tumour cell population. The term $aT\left(1 - \frac{T}{K}\right)$ represents logistic growth dynamics with intrinsic growth rate a and carrying capacity K in the absence of effector cells and TGF-β. The term $\frac{a_a ET}{g_2+T}$ is the number of tumour cells killed by effector cells. The parameter a_a measures the strength of the immune response to tumour cells. The third term $\frac{p_2 ST}{g_3+S}$ accounts for the increased growth of tumour cells in the presence of TGF-β. p_2 is the maximum rate of increased proliferation and g_3 is the half-saturation constant, which indicates a limited response of tumour cells to this growth-stimulatory cytokine [1].

$$\frac{dI}{dt} = \frac{p_3 ET}{(g_4 + T)(1 + \alpha S)} - \mu_2 I \tag{5.21}$$

The kinetics of IL-2 are described in equation 5.21. The first term $\frac{p_3 ET}{(g_4+T)(1+\alpha S)}$ represents IL-2 production which reaches a maximal rate of p_3 in the presence of effector cells stimulated by their interaction with the tumour cells. In the absence of TGF-β, this is a self-limiting process with half-saturation constant g_4 [1]. The presence of TGF-β inhibits IL-2 production, where the parameter α is a measure of inhibition. Finally, $\mu_2 I$ represents the loss of IL-2.

$$\frac{dS}{dt} = \frac{p_4 T^2}{\theta^2 + T^2} - \mu_3 S \tag{5.22}$$

Equation 5.22 describes the rate of change of the suppressor cytokine, TGF-β. According to [1], *experimental evidence suggests that TGF-β is produced in very small amounts when tumours are small enough to receive ample nutrient from the surrounding tissue. However, as the tumour population grows sufficiently large, tumour cells suffer from a lack of oxygen and begin to produce TGF-β in order to stimulate angiogenesis and to*

evade the immune response once tumour growth resumes. This switch in TGF-β production is modelled by term $\frac{p_4 T^2}{\theta^2 + T^2}$, where p_4 is the maximum rate of TGF-β production and τ is the critical tumour cell population in which the switch occurs. The decay rate of TGF-β is represented by the term $\mu_3 S$.

5.4.2 The System Dynamics Model

Model Stocks, Flows and Information

The SD model contains four stock variables, tumour cells, effector cells, IL-2 and TGF-β, as shown in Figure 5.31:

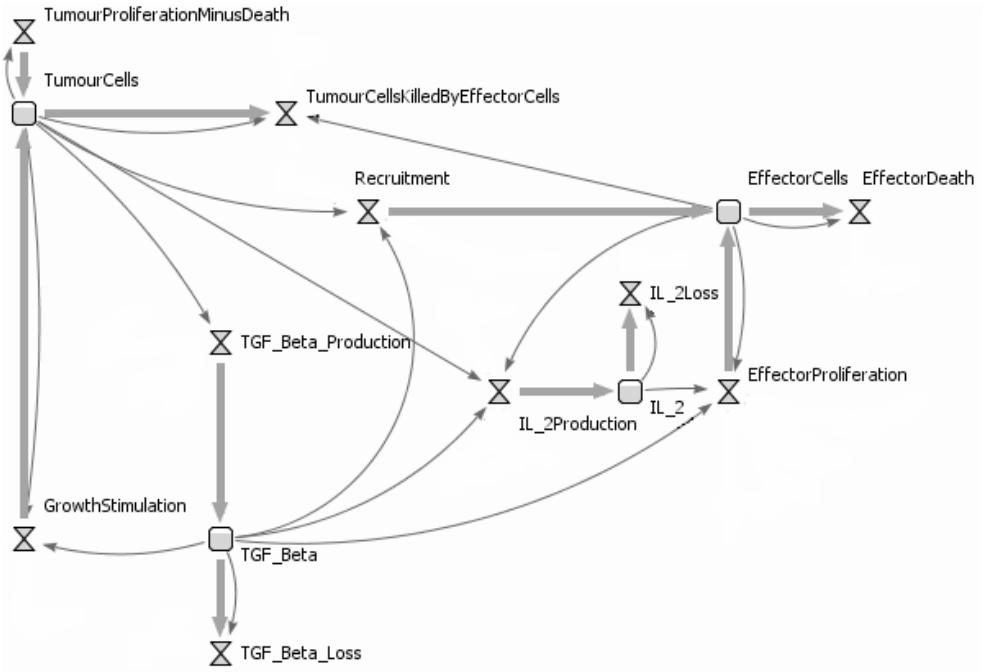


Figure 5.31: Stocks, flows and information for case 6

The stock of effector cells is changed by the recruitment of new effector cells (according to the number of tumour cells and TGF-β), death and proliferation. There is information from the effector cells stock to proliferation and death flows, as shown in Figure 5.31 above. The tumour cell stock is changed by natural proliferation and death, by the number of tumour cells killed by effector cells and by growth stimulation by TGF-β.

IL-2 stock changes with the production of new IL-2 molecules from effector cells (the production also depends on the number of tumour cells and TGF-β) and loss, which is proportional to IL-2 quantities in the system. TGF-β stock changes with production, which depends on the number of tumour cells, and loss.

Model Parameters

The model parameters are the same as those from the mathematical model, i.e. c, $gamma$, $mu1$, $p1$, $g1$, $q1$, $q2$, a, $g2$, $p2$, $g3$, $mu2$, $alpha$, $g4$, $p3$, $p4$, $theta$ and $mu3$.

Flows Calculations

Table 5.14 shows how the equations from the mathematical models are defined for the flow values:

Table 5.14: Flow calculations for case 6

Stock	Flow	Expression	Flow formula
EffectorCells	$EffectorProliferation$	$\left(\frac{p_1 EI}{g_1+I}\right) \cdot \left(p_1 - \frac{q_1 S}{q_2+S}\right)$	$\frac{p_1.IL\_2.EffectorCells}{g1+IL\_2} \cdot \left(p1 - \frac{q1.TGF\_Beta}{q2+TGF\_Beta}\right)$
	$EffectorDeath$	$\mu_1 E$	$mu1.EffectorCells$
	$Recruitment$	$\frac{cT}{1+\gamma S}$	$\frac{c.TumourCells}{1+gamma.TGF\_Beta}$
TumourCells	$TumourProliferation-MinusDeath$	$aT\left(1 - \frac{T}{K}\right)$	$TumourCells.a.\left(1 - \frac{TumourCells}{1000000000}\right)$
	$TumourCellsKilled-ByEffectorCells$	$\frac{a_a ET}{g_2+T}$	$\frac{aa.TumourCells.EffectorCells}{g2+TumourCells}$
	$GrowthStimulation$	$\frac{p_2 ST}{g_3+S}$	$\frac{p2.TGF\_Beta.TumourCells}{g3+TGF\_Beta}$
IL_2	$IL\_2Production$	$\frac{p_3 ET}{(g_4+T)(1+\alpha S)}$	$\frac{p3.EffectorCells.TumourCells}{g4+TumourCells} \cdot \frac{1}{(1+alpha.TGF\_Beta)}$
	IL_2Loss	$\mu_2 I$	$mu2.IL\_2$
TGF_Beta	$TGF\_Beta\_Production$	$\frac{p_4 T^2}{\theta^2+T^2}$	$\frac{p4.TumourCells^2}{theta^2+TumourCells^2}$
	TGF_Beta_Loss	$\mu_3 S$	$mu3 \times TGF\_Beta$

The Final System Dynamics Model

The final SD stock and flow diagram is depicted in Figure 5.32:

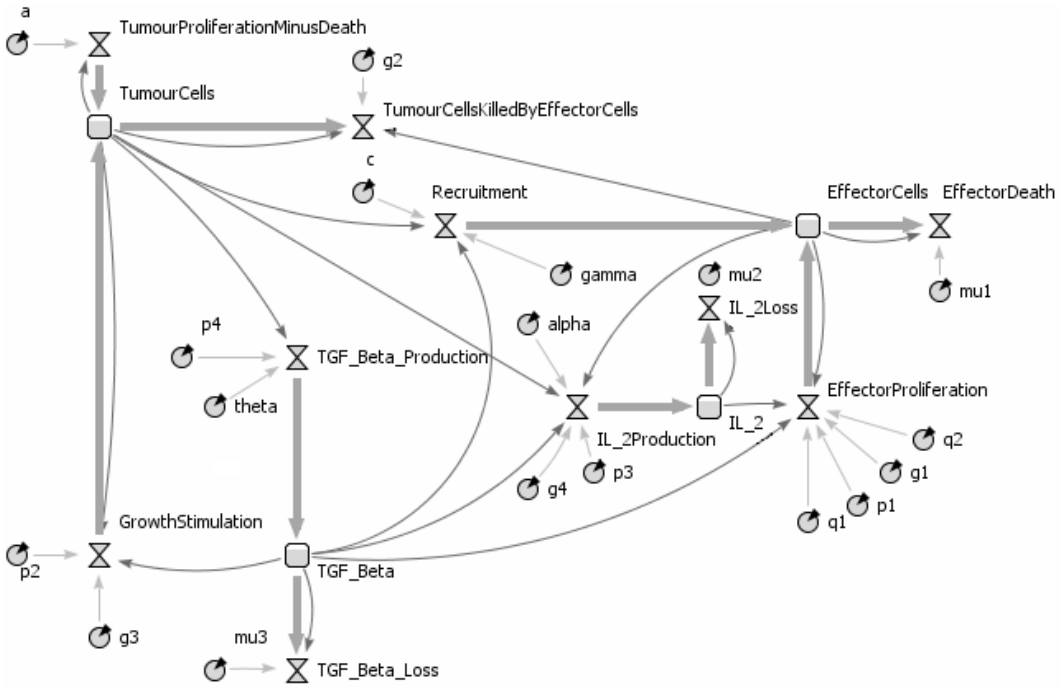


Figure 5.32: SD model for case 6

5.4.3 From System Dynamics to Agent-based Modelling and Simulation

Model Agents

Our agents correspond to the stocks in the SD model, the populations of agents therefore being the effector cells, tumour cells, IL-2 and TGF-β.

Agents Behaviours

The behaviour of each agent is shown in Table 5.15 below:

Table 5.15: Agents' parameters and behaviours for case 6

Agent	Parameters	Reactive behaviour	Proactive behaviour
Effector Cell	$mu1$	Dies	
	$p1$, $g1$, $q1$ and $q2$		Reproduces
	c	Is recruited	
	aa and $g2$		Kills tumour cells
Tumour Cell	a	Dies	
	a		Proliferates
	aa and $g2$	Dies killed by effector cells	
	$g3$ and $p2$	Has growth stimulated	
	$p4$ and $tetha$		Produces TGF-β
	c		Induces effector recruitment
IL-2	$alpha$, $p3$ and $g4$	Is produced	
	$mu2$	Is lost	
TGF-β	$p4$ and $tetha$	Is produced	
	$mu3$	Is lost	
	$p2$ and $g3$		Stimulates tumour growth

Agent Implementation

Based on the agents, parameters and behaviours derived from the previous step, state charts for each agent type were developed, as illustrated in Figure 5.33.

The ABMS model rates corresponding to the flow values in the SD model are given in Table 5.17. In the transition rate calculations, the variable $TotalTumour$ corresponds to the total number of tumour cell agents; the variable $TotalEffector$ is the total number of effector cell agents, $TotalIL\_2$ is the total number of IL-2 agents and $TotalTGFBeta$ is the total TGF-β agents.

5.4.4 Experiment

The experiment was conducted assuming the parameters of Table 5.16.

5.4.5 Results

Results for case 6 SD and ABMS simulations are provided in Figures 5.34, 5.35, 5.36 and 5.37. For all experiments, ABMS demanded far more computational resources than the SD simulation runs. Results demonstrate that the behaviour of the curves for effector

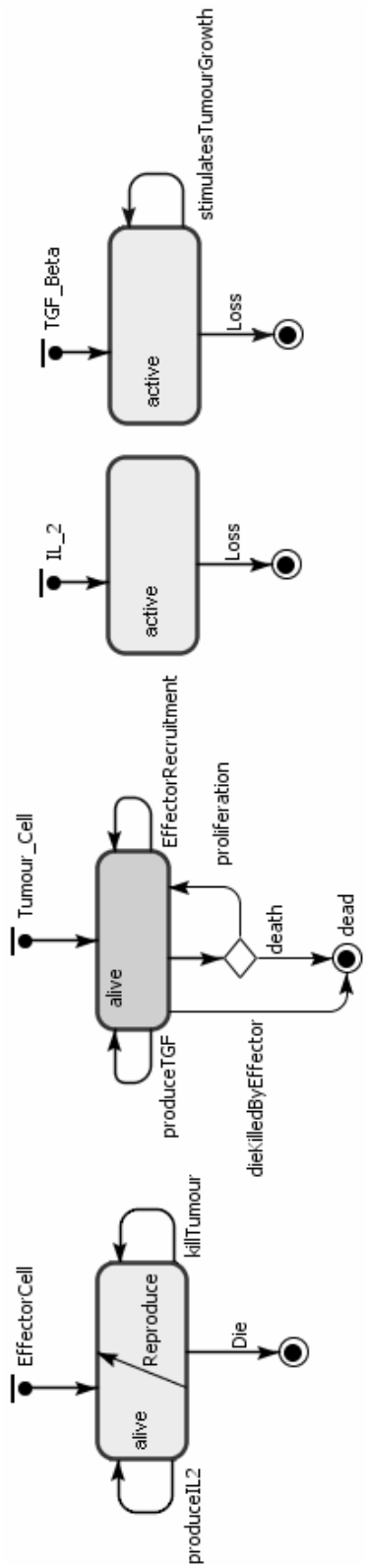


Figure 5.33: ABS state charts for the agents of case 6

Table 5.16: Parameter values for case 6

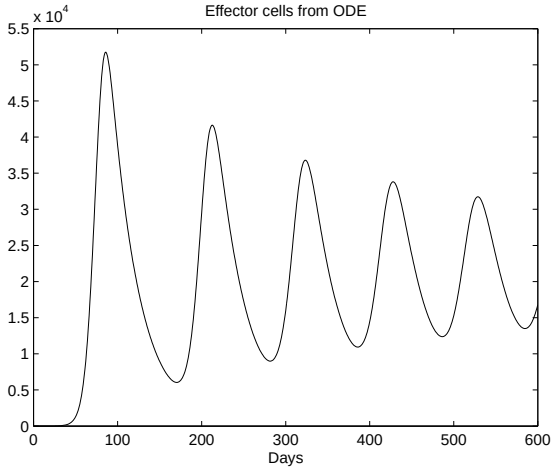
Parameter	Value
a	0.18
aa	1
alpha	0.001
c	0.035
g1	20000000
g2	100000
g3	20000000
g4	1000
gamma	10
mu1	0.03
mu2	10
mu3	10
p1	0.1245
p2	0.27
p3	5
p4	2.84
q1	10
q2	0.1121
theta	1000000

cells, tumour cells and IL-2 in both paradigms is similar, although the starting time for the growth of populations for the ABMS varies for each run. In the figures corresponding to the ABMS results, therefore, ten distinct runs were plotted to illustrate the variations. For most ABMS runs the pattern of behaviour of the agents is the same as that obtained by the SD. However for a few runs the populations decreased to zero, indicating that it is not always possible to obtain similar results with both approaches. Furthermore, the unexpected pattern obtained with ABMS should be further investigated by specialists to determine if it is realistic. In addition, the SD results for TGF-β reveal numbers smaller than one, which is not possible to achieve with the ABMS. That is the reason why there is no ABMS outcomes in Figure 5.37. The results for the simulations regarding these molecules are therefore completely different and the ABMS results are always zero.

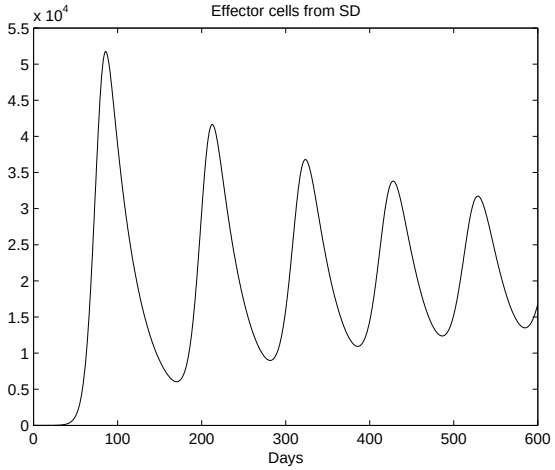
Figures 5.38, 5.39, 5.40 and 5.41 contrast the SD results with the closest results obtained from ABMS.

Table 5.17: Transition rates calculations from SD flows equations for case 6

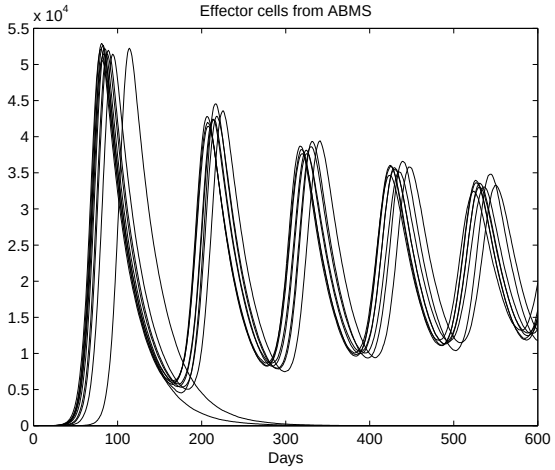
Agent	Transition	SD Flow equation	Transition rate
Effector Cell	Reproduce	$\frac{p_1 \times I \times EffectorCells}{g1+I} \times \left(p1 - \frac{q1 \times TGF\_Beta}{q2+TGF\_Beta}\right)$	$\frac{p_1 \times TotalIL\_2}{g1+TotalIL\_2} \times \left(p1 - \frac{q1 \times TotalTGFBeta}{q2+TotalTGFBeta}\right)$
	Die	$EffectorCells \times mu1$	$mu1$
	ProduceIL2	$\frac{p3.TumourCells.EffectorCells}{(g4+TumourCells)(1+alpha.TGF\_Beta)}$	$\frac{p3.TotalTumour}{(g4+TotalTumour)(1+alpha.TotalTGF)}$
	KillTumour	$\frac{aa \times TumourCells \times EffectorCells}{g2+TumourCells}$	$\frac{aa \times TotalTumour \times TotalEffector}{g2+TotalTumour}$
Tumour Cell	Reproduce	$\left(TumourCells.a\left(1 - \frac{TumourCells}{1000000000}\right)\right)$	$\left(TotalTumour.a\left(1 - \frac{TotalTumour}{1000000000}\right)\right)$
	Die	$\left(TumourCells.a\left(1 - \frac{TumourCells}{1000000000}\right)\right)$	$\left(TotalTumour.a\left(1 - \frac{TotalTumour}{1000000000}\right)\right)$
	DieKilledByEffector	$\frac{aa.TumourCells.EffectorCells}{g2+TumourCells}$	message from effector
	ProduceTGF	$\frac{p4.TumourCells^2}{teta^2+TumourCells^2}$	$\frac{p4.TumourCells}{teta^2+TumourCells^2}$
	EffectorRecruitment	$\frac{c.TumourCells}{1+gamma.TGF_Beta}$	$\frac{c}{1+gamma.TotaltGF}$
IL-2	Loss	$IL\_2.mu2$	$mu2$
TGF-β	Loss	$TGF\_Beta.mu3$	$mu3$
	stimulates TumourGrowth	$\frac{p2.TumourCells}{g3+TGF\_Beta}$	$\frac{p2.TotalTGF}{g3+TotalTGF}$



(a) ODE

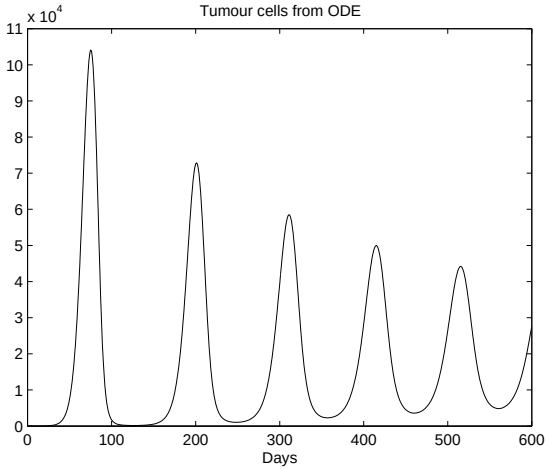


(b) SD

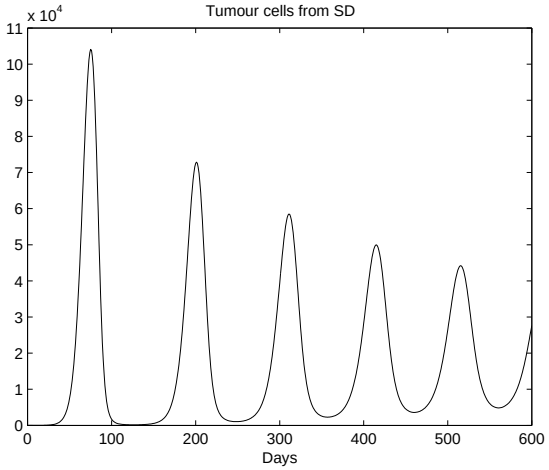


(c) ABMS

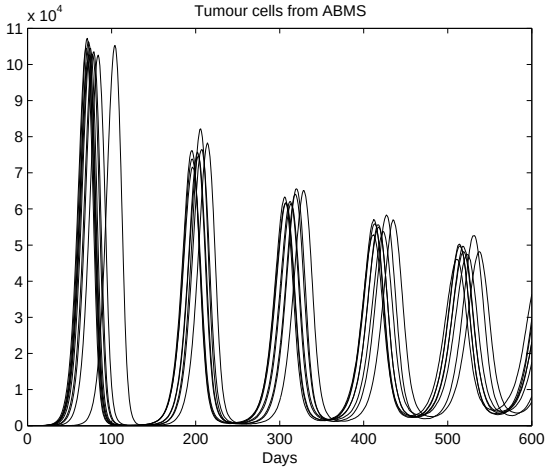
Figure 5.34: ODE, SD and ten runs of ABMS results for effector cells



(a) ODE

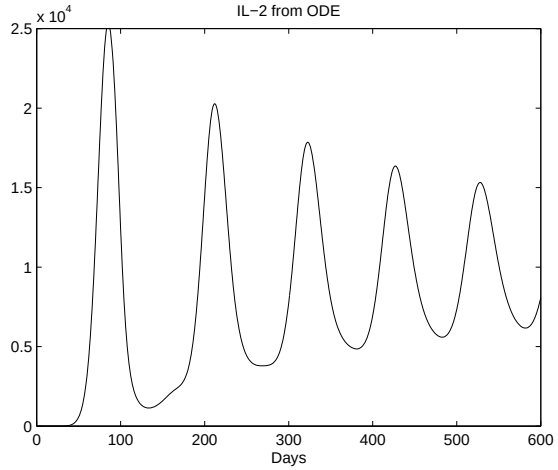


(b) SD

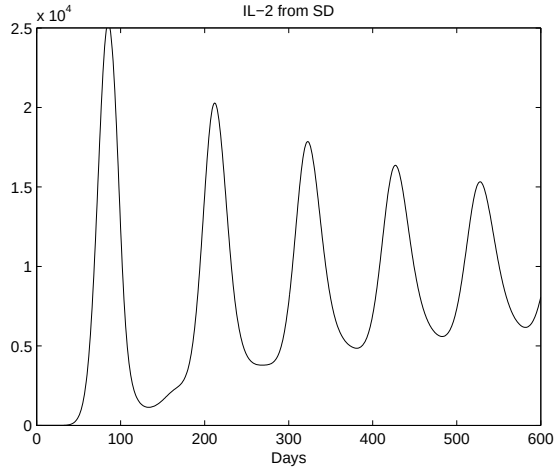


(c) ABMS

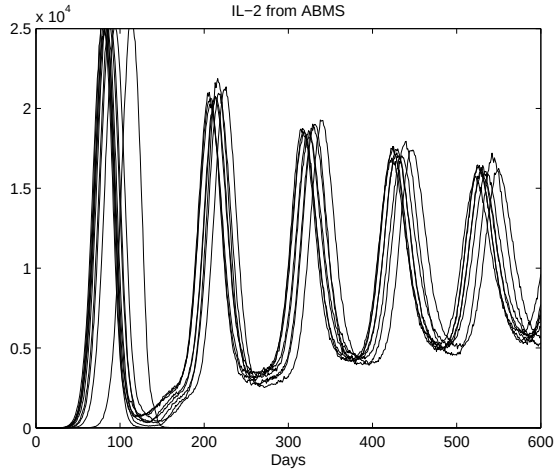
Figure 5.35: ODE, SD and ten runs of ABMS results for tumour cells



(a) ODE

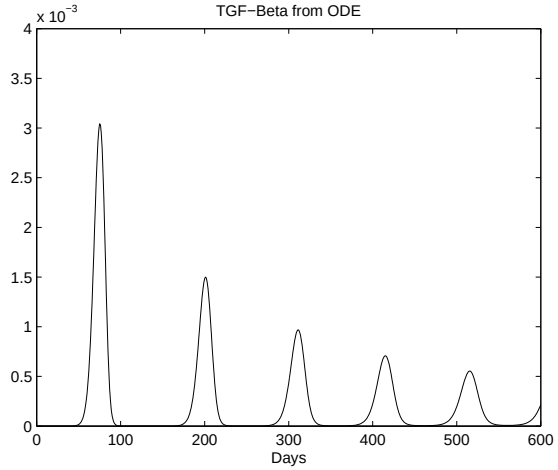


(b) SD

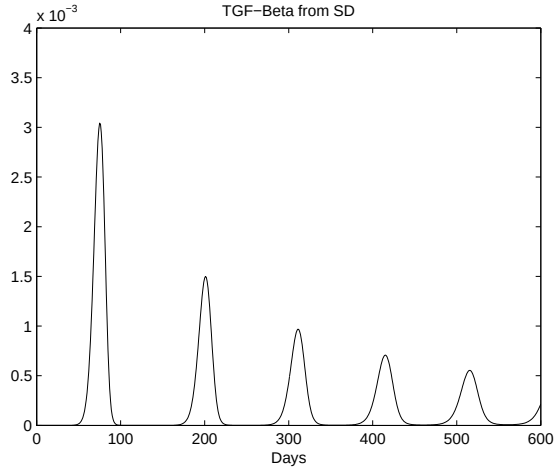


(c) ABMS

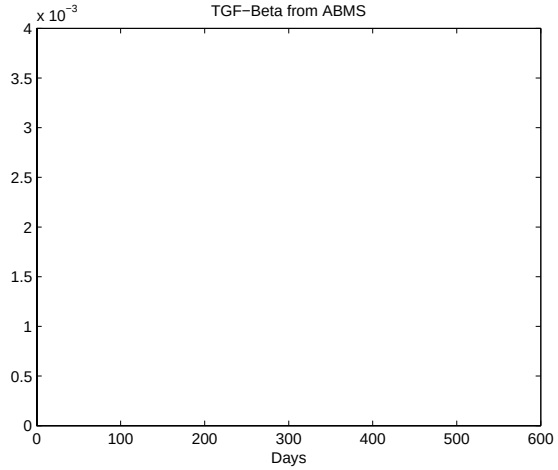
Figure 5.36: ODE, SD and ten runs of ABMS results for IL-2



(a) ODE

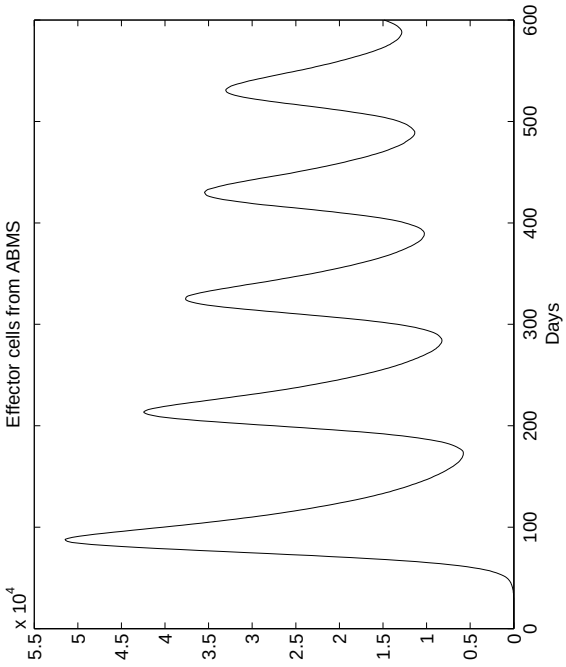


(b) SD

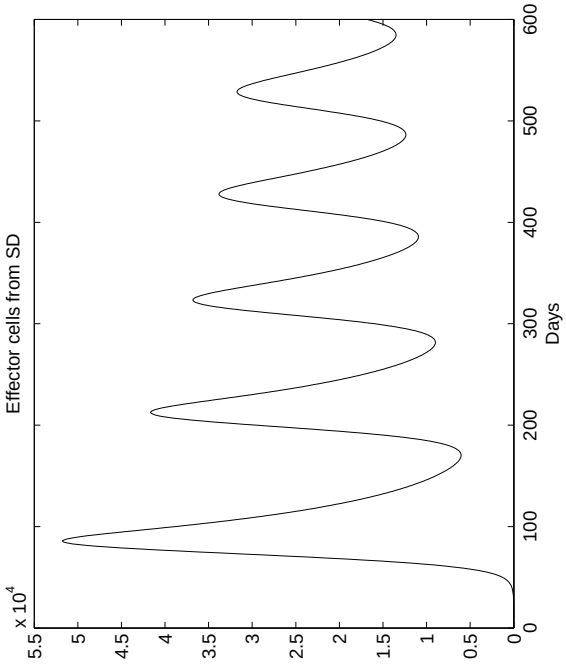


(c) ABMS

Figure 5.37: ODE, SD and ten runs of ABMS results for TGF-β

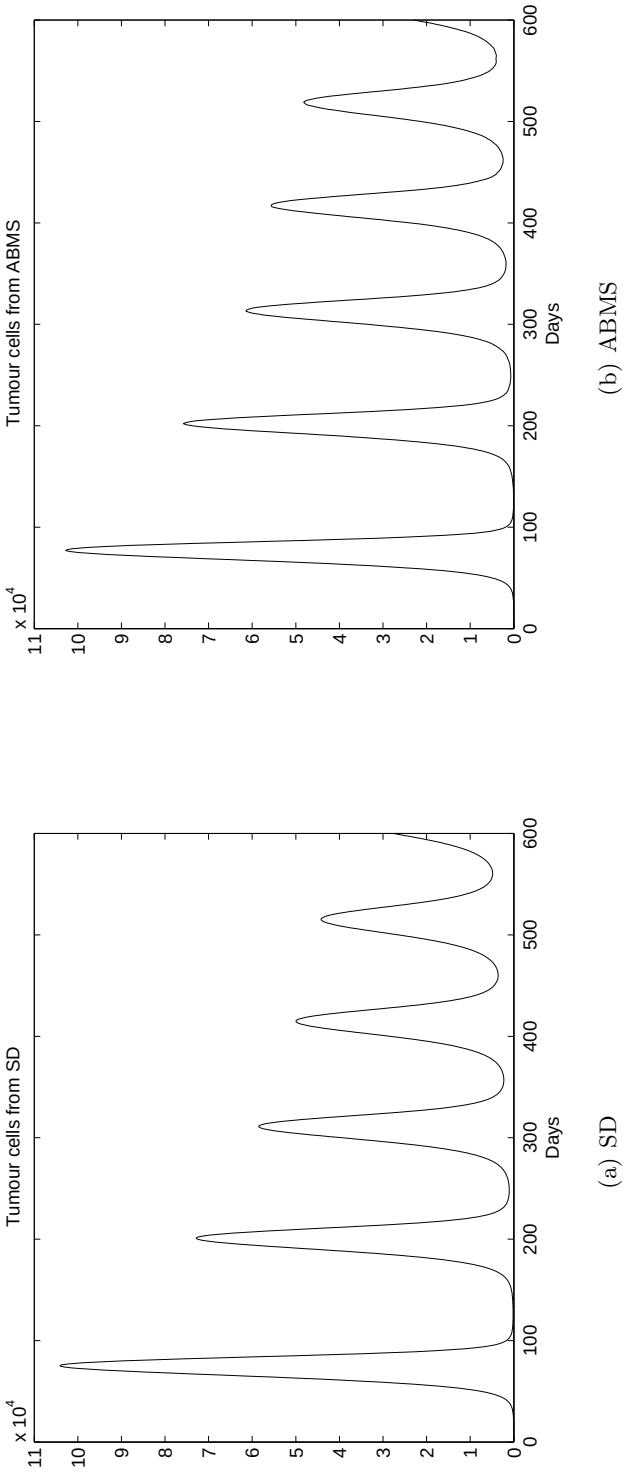


(e) ABMS



(d) SD

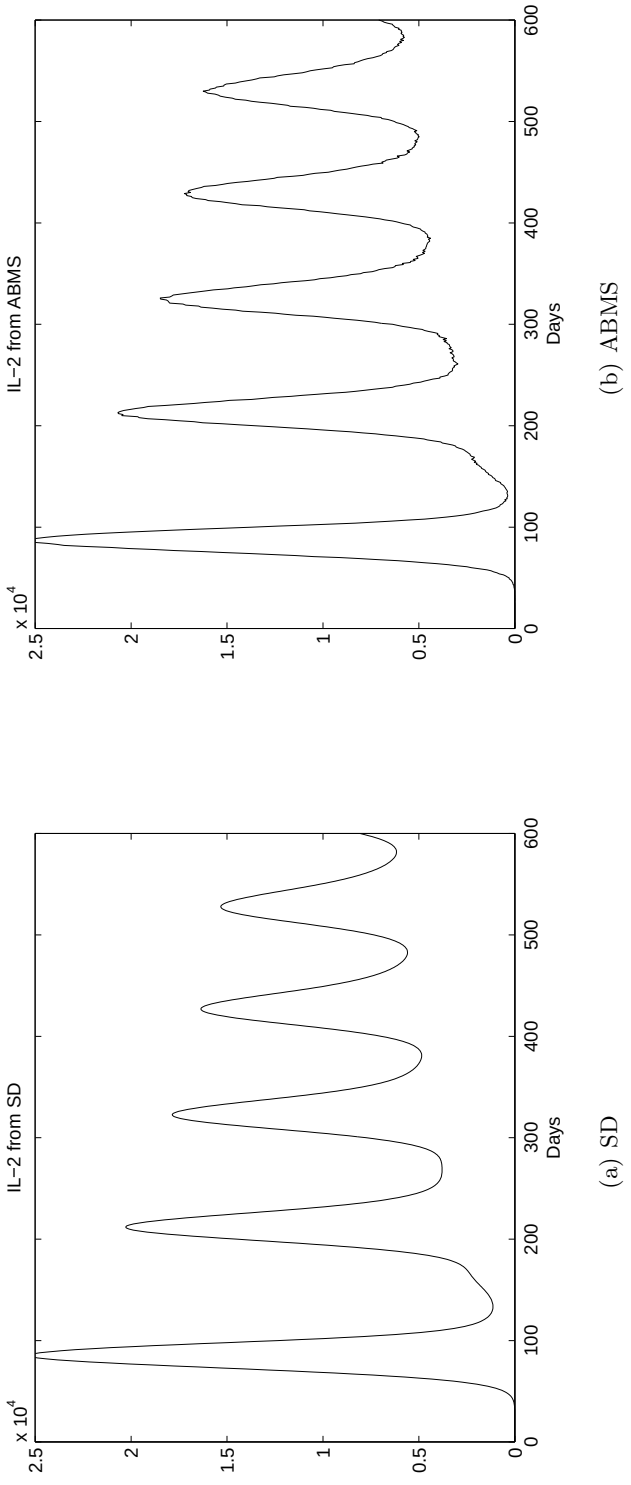
Figure 5.38: SD and ABMS results for effector cells



(a) SD

(b) ABMS

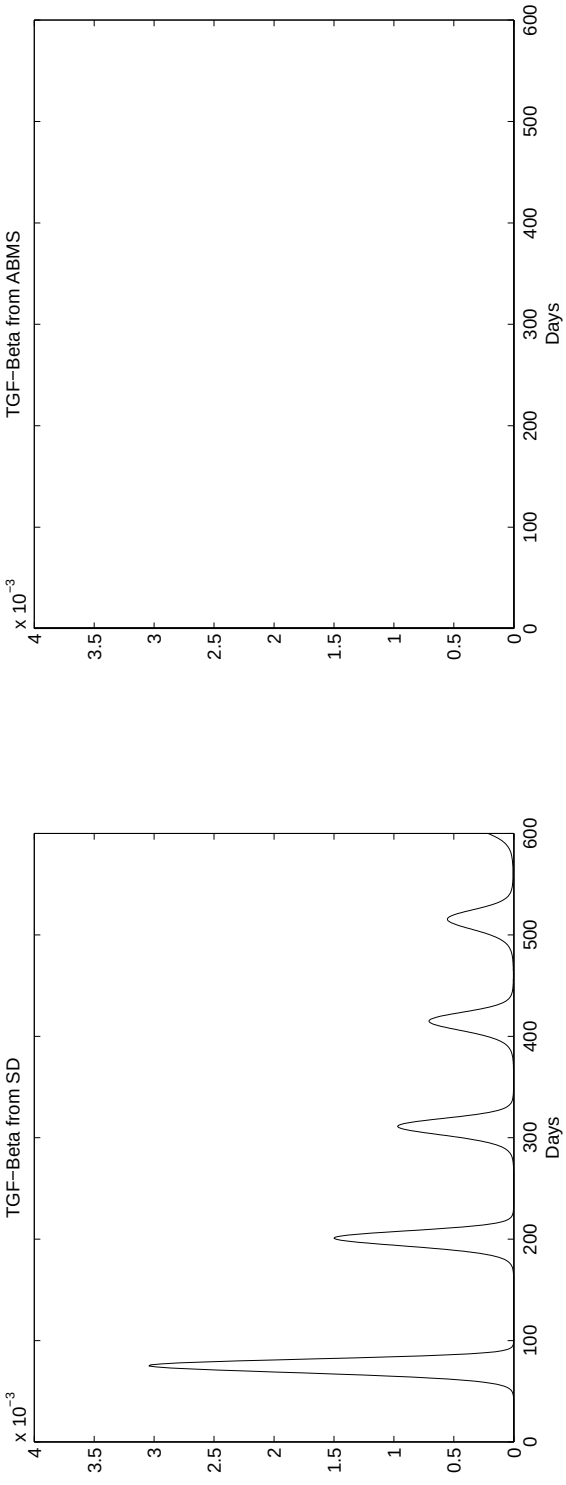
Figure 5.39: SD and ABMS results for tumour cells



(a) SD

(b) ABMS

Figure 5.40: SD and ABMS results for IL-2



(a) SD (b) ABMS

Figure 5.41: SD and ABMS results for TGF-β

5.4.6 Summary of Case 6 – Converting from System Dynamics to Agent-based Modelling and Simulation

In the previous sections we tested our conversion guidelines in a static four equation ODE model describing the interactions between tumour cells, effector cells, IL-2 and TGF-β molecules. The SD results matched those obtained in the ODE model, and in most cases the ABMS results resembled those from SD. For each different ABMS simulation run where results resembled, however, the growth and decay of tumour cells and effector cells had a slightly different starting point, due to ABMS randomness. Furthermore, there were some cases where ABMS produced another pattern of how the populations evolved over time: effector cells grew and eliminated all tumour cells before a period of two hundred days. In addition, ABMS was incapable to represent the TGF-β molecules dynamics, as their numbers in the SD and ODE models were smaller than zero.

By observing the results obtained, the conclusion is that for this case study it is not always possible to use both methodologies interchangeably. Moreover, in order to determine which simulation approach would be more suitable, further investigation using experimental data is necessary.

5.4.7 From Agent-based Modelling and Simulation to System Dynamics

In the ABMS scenario considered in this section, an effector cell E_{c_i} has to move towards a tumour cell T_{c_i} and kill it. The remaining effector cells in the population therefore will not have any impact on the death of T_{c_i}. Furthermore, an effector cell E_{c_j} only proliferates when it gets in touch with an IL-2 molecule, which moves towards it. In addition, when an effector cell E_{c_k} binds to a TGF-β molecule, it stops killing tumour cells and enters a sleeping state. The sleeping state remains until the effector cell binds another IL-2 molecule.

The Agent-based Model

The agents' state charts of the dynamic model are the same as those considered in the static model (Figure 5.33 of Section 5.4.3). The agents' behaviours defined in the static model (Table 5.15) remain. In addition, the random movement behaviour is considered for effector cell, tumour cell and IL-2 agents. The movement of the agents is controlled by events that occur in a certain rate (in our simulations, 0.01 was the rate for effector cell, IL-2 and TGFβ movement and 0.0001 for tumour cell movement). At each step of the simulation, an effector cell agent will look for a tumour cell and move towards it in order to kill it. In addition, an IL-2 will move towards an effector cell to allow for its replication, or takes it out of the sleep state, depending on the current state of the effector cell. In order to do the killing, an effector cell will send a message to the corresponding tumour cell agent, which will then die. A TGFβ molecule moves towards an effector cell and puts it to sleep. The transition $dieKilledByEffector$ will now therefore be triggered by this message. In addition, the proliferation rate of the effector cells will be greater than zero only when this cell meets the IL-2 (distance between them equal to zero).

System Dynamics Model

The corresponding SD model is shown in Figure 5.42 below. Similar to the dynamic SD model for case 6, the effector cell population is split in three different populations (stocks): one that considers cells that are stimulated by IL-2 to reproduce or are in the sleeping state ($EffectorCellsInProliferation$), the stock which regards effector cells without proliferation ($EffectorCellsNoProliferation$) and a stock of effector cells in the sleeping state ($EffectorSleeping$). Elements from the stock *EffectorCellsNoProliferation* migrate to the stock $EffectorCellsInProliferation$ via flow $MeetIL_2$ according to the the minimal element of $[EffectorCellsNoProliferation, IL - 2]$. Similarly, the flow calculation from $EffectorSleeping$ to $EffectorCellsInProliferation$ is the minimal element of the minimal element of $[EffectorCellsNoProliferation, IL - 2]$).

Experiment

The parameter values considered were the same as those from the static model. The simulations were run for a period equivalent to four hundred days using both approaches. Fifty replications for the ABMS were run and the mean values for the outputs were collected.

Results

Experimental results are shown in Figures 5.43 to 5.46 below. For tumour and effector cells outcomes, the overall shape of the simulation curves for both approaches is very similar. For the ABMS results, however, the growth of these two populations is more accentuated. The trend curve for IL-2 is not the same, as the growth and trend lines for both curves are very distinct from each other. TGF-β decays in both simulations with different velocity.

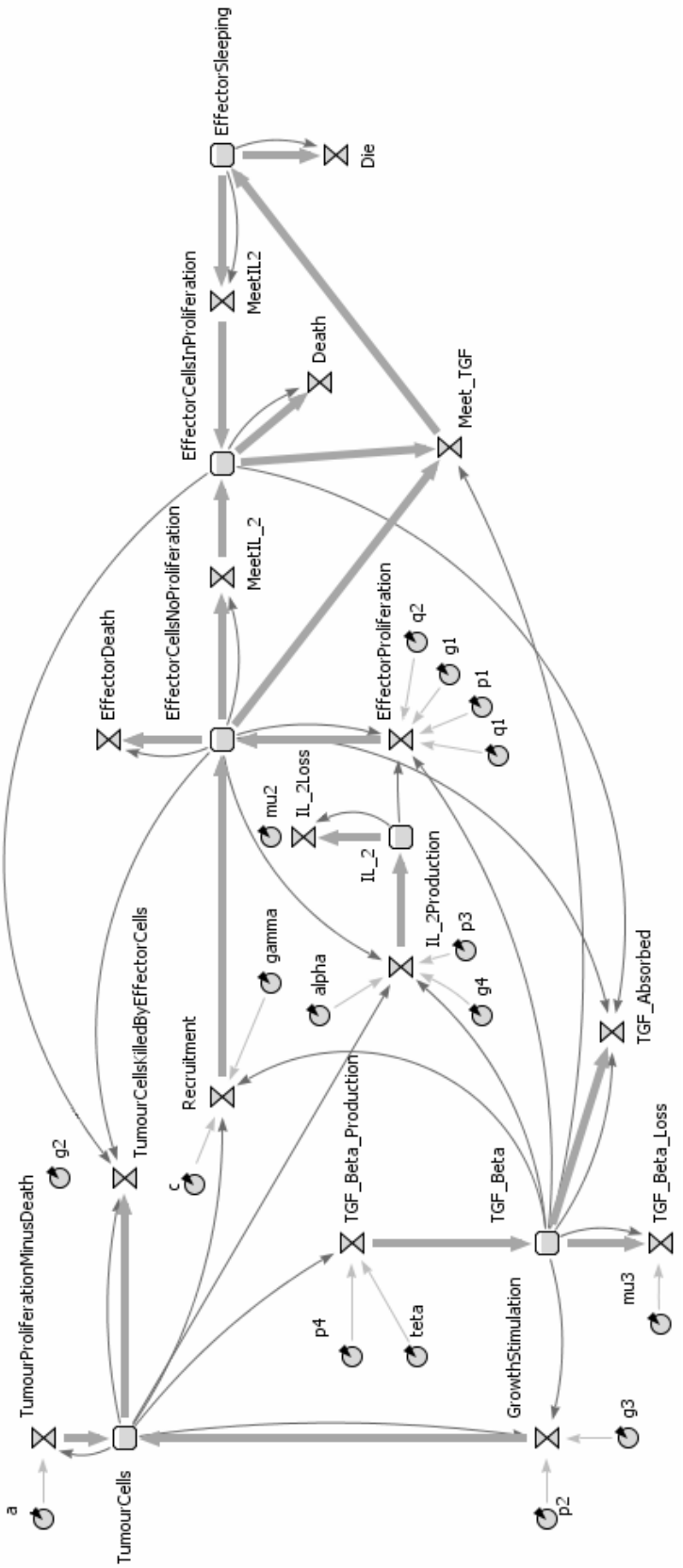
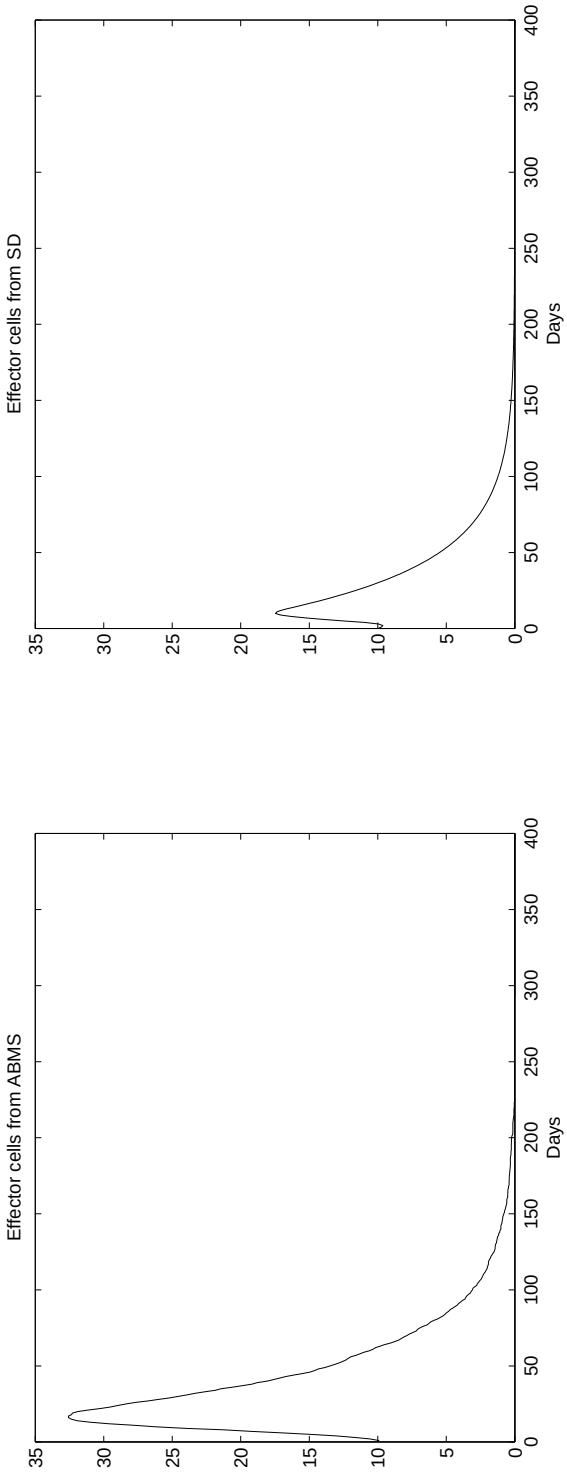


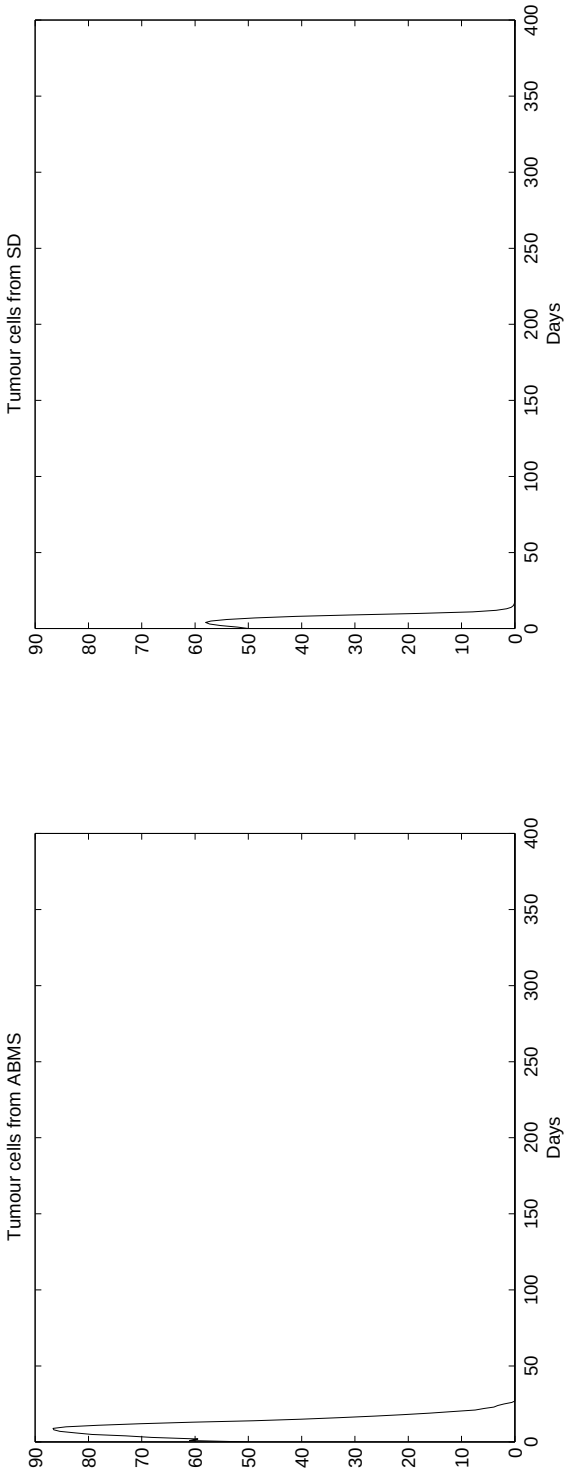
Figure 5.42: SD model corresponding to the dynamic agent-based model for case 6



(a) ABMS

(b) SD

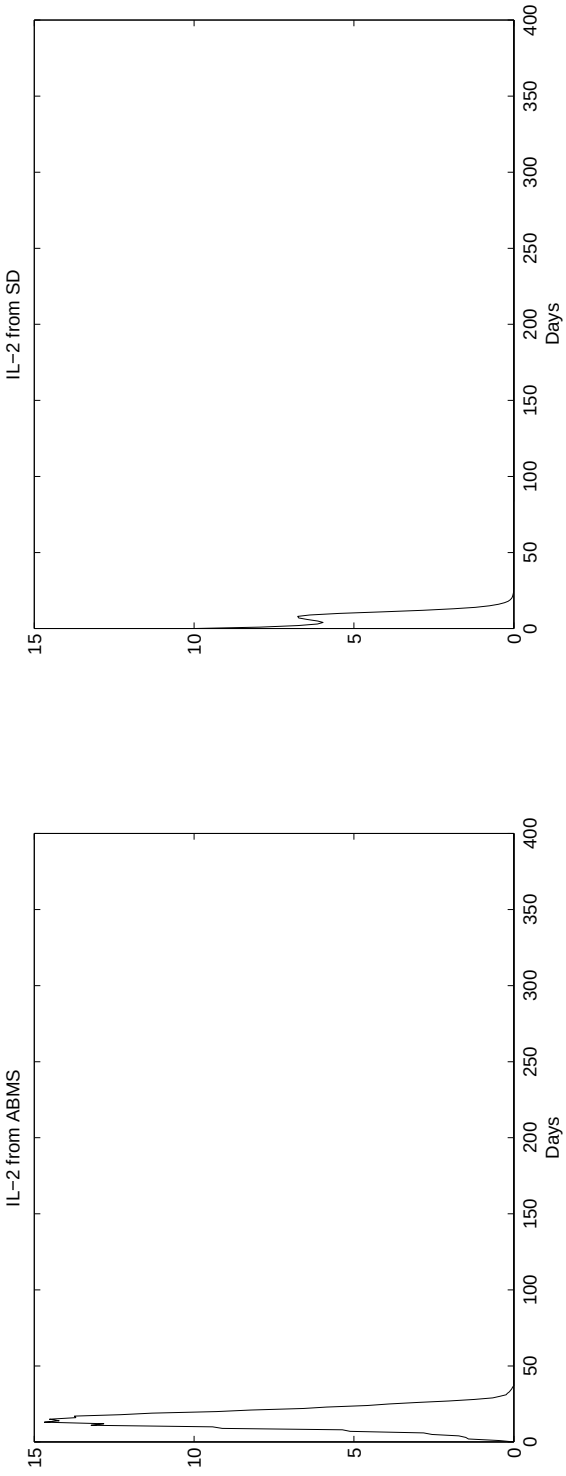
Figure 5.43: Results for effector cells considering cellular movement for case 6



(a) ABMS

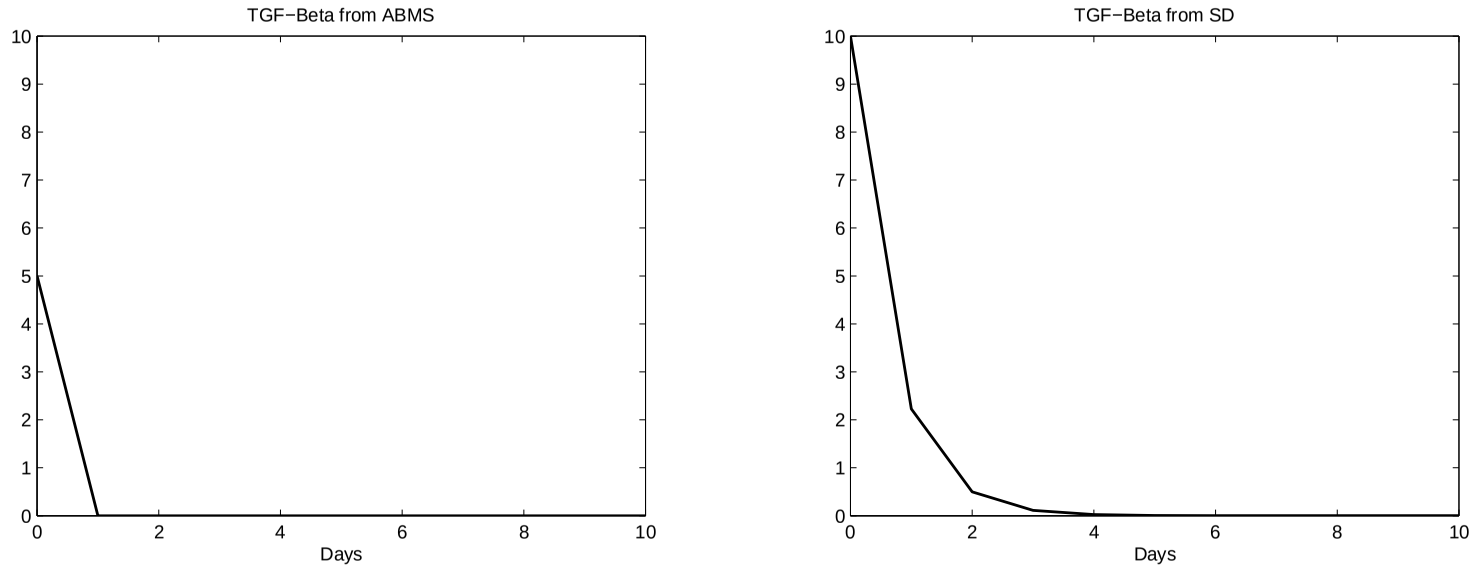
(b) SD

Figure 5.44: Results for tumour cells considering cellular movement for case 6



(a) ABMS (b) SD

Figure 5.45: Results for IL-2 considering cellular movement for case 6



(a) ABMS

(b) SD

Figure 5.46: Results for TGF-β considering cellular movement for case 6. As ABMS is discrete and the values for the TGF-β in the model are smaller than zero, they do not appear in the outcome.

Summary of Case 6 – Converting from Agent-based Modelling and Simulation to System Dynamics

In Section 5.4.7 a scenario involving interactions between tumour cells, effector cells, cytokines IL-2 and TGF-β was considered. In addition, the movement of these cells and molecules was implemented in the simulations. As in the previous case study, the objective was to build an ABMS simulation implementing the agents movement and subsequently converting this model into an SD model. The guidelines to convert from ABMS were used, and the outcomes from the obtained SD simulation revealed that, for effector cells, tumour cells and TGF-β it was possible to obtain similar patterns of the populations dynamics; however, the quantities of individuals for both approaches were different, which suggests that further calibration of the SD model might be necessary. In addition, SD was not able to mimic the erratic behaviour observed in the IL-2 population and therefore the outcomes were very distinct.

5.5 Summary

In this chapter three more case studies were presented in order to evaluate the conversion between approaches and compare the results obtained. The case studies considered established non-spatial mathematical models from literature describing interactions between the immune system cells and molecules, and tumour cells. Each mathematical model was first converted into an SD model and, subsequently, into an ABMS model. The results of this were then evaluated. Furthermore, this chapter experimentation differ from that from the previous chapter as it also tests the conversion from ABMS to SD. In addition, the ABMS models were modified to consider spatial movement of individual entities in the immune and tumour populations with the objective to verify the possibility to obtain an equivalent SD model.

The fourth case study was concerned with the use of ODEs to model interactions with general immune effector cells and tumour cells. The objective of this model is to observe

these two population evolution overtime and evaluate the impacts of cancer treatment in their dynamics. Four different scenarios regarding distinct sets of parameters were investigated and in the first three treatment was included. The conversion from ODEs to SD guidelines allowed for the construction of an equivalent SD model with the same outcomes as those obtained by the mathematical model. The equivalent ABMS resulting from the conversion, however, obtained very distinct results for most scenarios. The outcomes from SD and ABMS only resembled for scenario one, although ABMS curves reflected its random characteristic. In order to achieve closer results in the remaining three scenarios, constraints that the population sizes should be greater than zero were therefore added, which improved the similarity of both approaches results for only two scenarios. It appears that two major characteristics of this model influenced the unsuccessful conversion: (1) The small quantities of individuals considered in the simulations – specially regarding the effector population size, which was always smaller than ten – that significantly increase the variability of the ABMS; and (2) The original mathematical model considers population sizes smaller than one, which was not possible to be achieved by the implemented ABMS. In addition to this particular model's characteristics, for any mathematical/SD model considering intervals of growth or decay of populations observed in our studies, the corresponding curves in the ABMS outcomes are more accentuated, given the fact that SD changes the stocks quantities continuously whereas ABMS varies discretely. These results confirm the findings obtained in the previous chapter, where the outcome differences were also due to the characteristics of each modelling approach.

Regarding the conversion from ABMS to SD for case four, the resulting SD model for the static ABMS produced the same results as those from the mathematical model, which indicates that the variability observed in the ABMS is not reflected in the SD. In order to further explore the effectiveness of our guidelines, we modified the static model to consider simple movements of the cells and molecules involved in the simulation model. Pertaining the dynamic model, differences were noticed regarding to the population sizes over time. This suggests that extra efforts such as parameters calibration would

be necessary to obtain SD results closer to the outcomes produced by the ABMS.

The fifth case study referred to the investigation of the interactions between effector cells, cytokines IL-2 and tumour cells, and only one scenario was considered. For this case, the conversion guidelines were effective in assisting the construction of both SD and ABMS simulations, which produced very similar results as those from the original mathematical model. As populations' sizes had a magnitude of 10^4 individuals, the ABMS variance in the outcomes was not evident, which contributed to the outcomes similarity. Regarding the conversion from ABMS to SD, there was the need to complement the guidelines to convert from ABMS to SD, as they originally did not consider cases where agents' behaviours changed with time. By modifying the conversion framework it was possible to obtain an SD model. Similarly to the previous case study, the SD results did not reflect the variance observed in the ABMS outcomes.

Case six added complexity to the previous case study by establishing a mathematical model including the influence of the cytokine TGF-β in the interactions between effector cells, cytokines IL-2 and tumour cells. The SD obtained with the conversion guidelines produced the same outcomes as those from the ODE model. The simulation outcomes for the ABMS were mostly following the same pattern as that produced by the SD; however there were some alternative outcomes where the patterns of behaviour demonstrated a total extermination of tumour cells by the first two hundred days. This indicates that for this case study the ABMS results are more informative, as they illustrate another set of possible dynamics that should be validated through further immune experimentation. Regarding the conversion from ABMS to SD, it was observed that due to the increase of elements and behaviours in the ABMS, the equivalent SD becomes very complex and less intelligible. In addition, there is also an increase in the model building effort. The obtained SD results do not resemble those from the ABMS, as they vary in quantities and do not regard the variability observed in the ABMS outcomes.

By observing our case studies and their outcomes, it was not possible to define a general framework that would definitively determine which approach was the most appropri-

ate for a certain problem. In our last case studies, however, we could observe some characteristics to be considered when choosing the approach:

- SD is incapable of reflecting exactly the same variability as that obtained from the agent-based simulation, as it is a deterministic approach.
- SD variables change continuously in time and therefore population growth and decay might be different from those obtained by the agent-based simulation.
- As the numbers of different agents and behaviours increase, the corresponding SD becomes very intricate and difficult to develop and understand.
- The previous item suggests that there are cases where it is preferable not to convert between approaches as the agent-based model is easier to conceptualise and implement.
- The verification and validation processes of a very complex SD model might not be trivial.
- SD is less informative than ABMS, as it does not produce multiple scenarios or variations over the course of more than one run within the same parameters.

Chapter 6

Conclusions

6.1 Introduction

The research presented in this thesis investigated methods of translating and obtaining similar simulation outcomes regarding a number of different simulation modelling approaches. The main motivations for this work is provided by the emerging research in multi-paradigm and multi-modelling simulations, as well as the importance of investigating the application of such methodologies and compare outcomes to aid advancing immunology research. Immunological phenomena belong to the area of complex systems, involving many interacting elements that can also adapt to further scenarios such as new infections. System simulation is therefore suitable for investigating immune processes, as most of its methods are closer to the natural description of the system in terms of representation of entities and their interactions. Further, immune simulation experimentation (1) avoids the extra costs involved in laboratory trials, (2) takes less time than real-world experimentation and (3) does not need to follow ethical protocols (Section 2.7).

Some immune problems can be implemented by more than one approach; when and whether these approaches provide equivalent outcomes is not fully understood. In addition, translating between approaches for immunology is important because research on

processes in the immune system constantly gathers new information. This requires the corresponding simulations to be updated frequently to suit the latest requirements. In some cases, the replacement of the current simulation approach for new developments also needs to be considered in order to meet these demands. Another possible advantage of the conversions guidelines would be in circumstances where a model developer is well acquainted with one simulation approach and would like to use his current models to learn another technique. In cases where there is a established model in a certain simulation approach, conversion techniques to translate the current model to another approach would be a good starting point to learn this last approach and possibly expand the model.

Mathematical models – in particular ODEs – are largely used in immune modelling; however, this paradigm presents limitations regarding emergent behaviour, individual interactions and an increase in problem complexity. SD is an alternative approach to model problems undertaken in numerical simulation within a system's thinking perspective. ABMS, as distinct from ODEs and SD, complements mathematics as (1) it provides a means to implement individuals from the system in a natural way, (2) it is used to explore emergence and (3) it represents individual memory and spatial localization.

In order to support the implementation of multi-paradigm simulations, as well as performing the replacement of a simulation approach, guidelines for the conversion between ODE models, SD models and ABMS models of the immune system were therefore developed as a research contribution in this thesis. These guidelines were tested, improved and their effectiveness assessed using case studies.

While answering the research question we also produced a framework based on that conceived by [70] for conducting simulation studies in immunology, and outlined the pitfalls that might be encountered during the development of a simulation model. This guidance was derived from a review of the immunology literature related to simulation. It defined general steps to be followed when developing an immune simulation and which encompass common aspects to be considered during the simulation development,

independent of the simulation approach adopted.

6.2 Evaluation of Aims

The central aim of this thesis was to develop, test and validate our own set of guidelines for converting between approaches: from ODE models to SD, from SD to ABMS, from ODE to ABMS and from ABMS to SD.

A secondary aim was to discuss the merits of SD and ABMS for immunology to assist researchers with their choice between both approaches, to determine if the simulation problems associated with the use of these modelling techniques are interchangeable and to establish those circumstances in which one approach would be preferable to the other.

To achieve these aims, the following objectives were defined:

1. To define and test guidelines to convert between (1) ODE models to SD models, (2) SD models to ABMS, (3) ODE models to ABMS and (4) ABMS to SD models and assess the impact of these conversions.

2. To compare outcomes considering aspects such as the behaviour of the entities of the model (whether they are static or interact with other entities and whether they have spatial representation or not); the type of hypothesis to be tested and the modelling effort.

3. To define guidances to assist immune simulation developers in choosing between approaches, depending on the characteristics of the problem to be undertaken.

Six case studies derived from ODE models of immune problems – involving immune system ageing, the spread of the HIV virus and interactions between the immune system and tumour cells – were selected to test our guidelines and achieve our objectives.

To meet the first objective, we introduced our guidelines in Section 3.5.1 to convert from ODEs to SD. These guidelines were tested against all six case studies (Chapter 4 and Chapter 5).

Objective two was fulfilled by defining the guidelines to convert from SD to ABMS in Section 3.5.2. In addition, by comparing the results, it was partly possible to attain the last goal, which was to provide guidance on the choice between approaches for the case studies. We also defined guidelines to convert from ODEs to ABMS (Section 3.5.3), which were very similar to the conversion from SD to ABMS. Our focus in this work, therefore, was on testing the conversion from SD to ABMS.

The third objective was achieved in Section 3.5.4, in which we introduced our conversion guidelines to translate from ABMS to SD.

As it was not possible to define a general framework that would definitively determine which approach was suitable, the last objective was only partially realised. There is, therefore, the need for further research regarding this topic, as we will explain in sections 6.4 and 6.5.

Experimentation

The experiments used to achieve our objectives were split in two sets: the first set comprised three case studies, which involved static models. The second batch included another three case studies with simple interacting agents and movement. For each case study, the mathematical models were converted into SD models and then into ABMS by following our guidelines. Additionally, in the second set of case studies, the guidelines to convert from ABMS to SD models were also tested.

The simulation outcomes were validated against the results produced originally by the mathematical models. In order to further compare the results produced by SD simulations and ABMS, the statistical Wilcoxon (Mann-Whitney) test was also selected. The comparison of results for both approaches was conducted in order to evaluate (1) if the approaches were equivalent (model verification), (2) if the results were similar, (3) if the approaches could be used interchangeably for a particular case study and (4) if one approach would be preferable than the other for the case study.

The outcome comparison was presented in Chapter 4 and Chapter 5, for all case stud-

ies. As the case studies comprised different scenarios with regard to modelling effort, interacting and non-interacting elements, and spatial and non-spatial localisation, it was possible to achieve the fourth objective.

6.3 Research Findings

For all case studies, the guidelines to convert from ODE to SD were effective and produced the same results as those from the mathematical model. Furthermore, it was also possible to obtain the respective ABMS, although differences on the outcomes occurred due to particularities of each approach. Some differences observed were not statistically relevant, however, there were cases where dissimilarities were noticeable. The key outcome differences are summarised in Table 6.1. When applicable, we suggest the preferable approach to be considered (on the fifth column of the table), which also contains information about the population size, as it appears that these influence the outcomes.

With regard to the conversion from ABMS to SD, based on our experimentation and findings (Table 6.2), we suggest some important considerations to be made before attempting to perform the translation:

- SD is incapable of reflecting exactly the same variability as that obtained from the agent-based simulation, as it is a deterministic approach.
- SD variables change continuously in time and therefore population growth and decay might be different from those obtained by the agent-based simulation.
- As the numbers of different agents and behaviours increase, the corresponding SD becomes very intricate and difficult to develop and understand.
- The previous item suggests that there are cases where it is preferable not to convert between approaches as the agent-based model is easier to conceptualise and implement.

Table 6.1: Summary of findings with regards to the conversion from SD to ABMS

Case Study	Outcome of the comparison	Explanation	Population sizes	Preferable approach	Section
1) Naive T cell output	• Results were statistically the same	• Large populations • Less variability in the agents' population	10^3	SD	4.2.7[i]
2) Tumour growth	• The growth curve was more accentuated for the agent-based approach • Agent-based is limited with regards to model scalability	• Agents grow discretely •Agents require more computational resources	Varied from 10 to 10^{70}	SD for large sizes	4.3.5[ii]
3) HIV spread	• Each run outcome produced slight variations with regards to the start of the HIV infection and spread	• Stochastic character of the agent-based models	10^4	Agent-based seems more realistic	4.4.5[iii]
4) Tumour/Effector	• Most results were different	• Agents grow discretely • Stochastic character of the agent-based models • It appears that variabilities in small populations have major impacts in the outcomes	Varied from 6 to 600		5.2.5[iv]
5) Tumour/Efector/IL-2	• Results were statistically the same	• Large populations • Less variability in the agents' populations	10^4	SD	5.3.5[v]
6) Tumour/Effector/ IL-2/ TGF-β	•Different Runs with outcome variations • Simulations produced alternative scenarios •The behaviour of the curves is less erratic for agents	• Agent-based stochasticity • New scenarios need further investigations to assess their feasibility • Large numbers of agents	10^4		5.4.5[vi]

[i] Page 97
[ii] Page 111
[iii] Page 128
[iv] Page 149
[v] Page 176
[vi] Page 193

Table 6.2: Summary of findings with regards to the conversion from ABMS to SD

Case Study	Outcome of the comparison	Explanation	Population sizes	Preferable approach	Section
4) Tumour/Effector *static*	• The outcomes were different • The outcomes were different	• SD does not replicate the erratic behaviour of the agent-based outcomes as it is a deterministic approach.	10^3		5.2.10
Tumour/Effector *dynamic*	• The outcome curves had the same trend but some different values	• Constant growth of stocks from SD versus discrete number of agents	500		5.2.11
5) Tumour/Effector/ IL-2/ *static*	• The results were statistically the same	• Large population • Less agent variability	10^4		5.3.6
Tumour/Effector/ IL-2 *dynamic*	• Similar results for effector cells • Different results for the remaining populations	• Small variability on the agents • continuous versus discrete	50		5.3.6
6) Tumour/Effector/ IL-2/ TGF-β *dynamic*	• Results were different	• continuous versus discrete • stocks assume values smaller than zero • SD does not replicate the erratic behaviour of the agent-based outcomes	100 10^3		5.4.7

[i] Page 165
[ii] Page 165
[iii] Page 180
[iv] Page 183
[v] Page 207

- The verification and validation processes of a very complex SD model might not be trivial.
- SD is less informative than ABMS, as it does not produce multiple scenarios or variations over the course of more than one run within the same parameters.

6.4 Limitations of the Study

Based on our experimentation outcomes, it is possible to conclude that the guidelines are effective in producing equivalent models for our case studies. In addition, by further analysing the comparison of the results, it has been found that there are predictable differences due to the characteristics of each simulation model. Our contribution, however, presents limitations with regard to the nature of the case studies chosen and the simulations validation processes; for some case studies, we could not determine the best approach to be used.

All case studies investigated in this thesis were derived from ODE models established in the literature. Established SD models or ABMS models were not included in our investigation because they are rare or unavailable. Compared with ODEs, there are few ABMS models (which are not cellular automata) and SD models with data provided in which we could perform our testing. Further experimentation is therefore needed to address case studies that have either SD or ABMS as their original model.

We also tried to develop our own SD simulations from scratch, based on data provided by immunologists from the Biomedical Sciences department of the University of Nottingham and the Nottingham City Hospital. However, the data provided was not suitable for the simulation model development because there was no information about how the elements in the system would evolve with time. It was concluded that it would be necessary to collect another set of specific data in the laboratory.

As discussed in Chapter 3, the process of validation ensures that the model is sufficiently accurate for the purpose at hand. For immunology, it is acknowledged that models are

not intended to be accurate for a number of reasons: (1) for some case studies there is no real-world data to use in comparison, (2) for other case studies there is insufficient data, (3) real-world data is inaccurate and (4) even if the data is accurate, the real-world data is only a sample, which in itself creates inaccuracy.

In most of our case studies (1) there was no data available to validate the models, and (2) as previously stated, the data provided by immunologists was unsuitable for building a simulation from scratch. This led us to consider the outcomes of the original ODE models as our main source of validation. By possessing the laboratory experimentation data related to the immune experiments, we should be able to gain further insights into the most effective approach to be used.

Another limitation is that most models involve few agents with simple interactions. It appears that, for immunology, simple models are satisfactory in developing an understanding of a certain process. However, to test and improve our guidelines further, models which contain more populations of agents are needed. In addition, only agents with random movement and synchronous actualisation of their parameters and states were considered. There is therefore a requirement to verify if asynchronous agents in a model would be suitable for the SD conversion.

6.5 Future Work

While the guidelines performed well in converting between approaches in our case studies, there is the need to perform further tests in established system dynamics or agent-based models for immunology, in order to fully assess their effectiveness. Furthermore, we intend to develop our own models from scratch and to test the translation techniques. These models will be based on new immune data regarding T cells aging that will be collected by immunologists from Nottingham City Hospital. The decision about the information to be collected in the laboratory was taken by us, together with immunologists, in order to ensure that the final data set collected would be suitable for

our experimentation and validation processes. We hope that, by further investigating the simulation outcomes with real data, we will also be able to acquire more knowledge regarding the appropriateness of each paradigm for the problem undertaken.

Working more closely with immunologists, there are also other research enquiries we seek to investigate related to their acceptance of simulation as a research tool. Although there are examples showing the success of simulation in aiding advances in immunology, this set of methodologies is still not popular. There are three major reasons for this: (1) such tools are not fathomable in the immunology research field; (2) although simulation is acknowledged, there is no existing knowledge of how to use them; and (3) there is insufficient trust in the results produced by simulation. Hence, our next objective is to outline the potential contribution of simulation methods to support immunological studies and to invite experts, including simulation developers and computer scientists, to develop solutions in this field. In order to do so, we plan to develop and validate our new simulations, together with immunologists from Nottingham City Hospital and to assess their understanding and reliance on the results obtained.

Furthermore, we intend to show immunologists the possibility of improving the current immune mathematical models by presenting our translation methods. In addition, it is intended to assess the guidelines' effectiveness when used by non-simulation experts. As another future research project, we wish to investigate and develop a decision framework that assists with the choice of a simulation approach according to the problem presented, as a consequence of the new study. There is also the need to find out which simulation modelling paradigm is better accepted by immunologists and if it is necessary to develop specific simulation tools using immunological terminology to aid immune simulation research.

In addition, by further analysing the comparison of results, it was found that there are differences to be expected due to the characteristics of each simulation model. Many of these differences are already known within the simulation community; however, it is not always possible to predict when some of these differences will occur. We intend to further

investigate why and how these differences happen by modelling new case studies and by systematically determining when phenomena such as variability and extra scenarios arise.

There are cases where the same simulation scenario implemented in agent-based simulation produces more than one solution due to specific constellations within the random number stream. It is necessary, therefore, to gain additional insights into why and how frequently these extreme cases occur. For example, we could count the appearance of these unusual cases (as a measure of system stability or robustness of the solution) when running the experiments $10,000$ times. This could assist immunologists in defining vaccination strategies and the appropriateness of cancer treatments by making them aware of the possible outcome scenarios and how frequently they take place.

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