A collaborative relationship between the Analytical Scientist and the Statistician

Making the right decision with data to give appropriate cut-points that can be applied for sample analysis

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Overview

This presentation will outline:

- Collaborative process in the context of updated recommendations for statistical analyses
- Importance of the analytical scientist's understanding of how the statistical analysis should be applied and it's appropriateness for the analytical data – post recommendation updates
- 3 case studies – illustration of importance of the collaborative relationship between Statistician and Scientist

Introduction

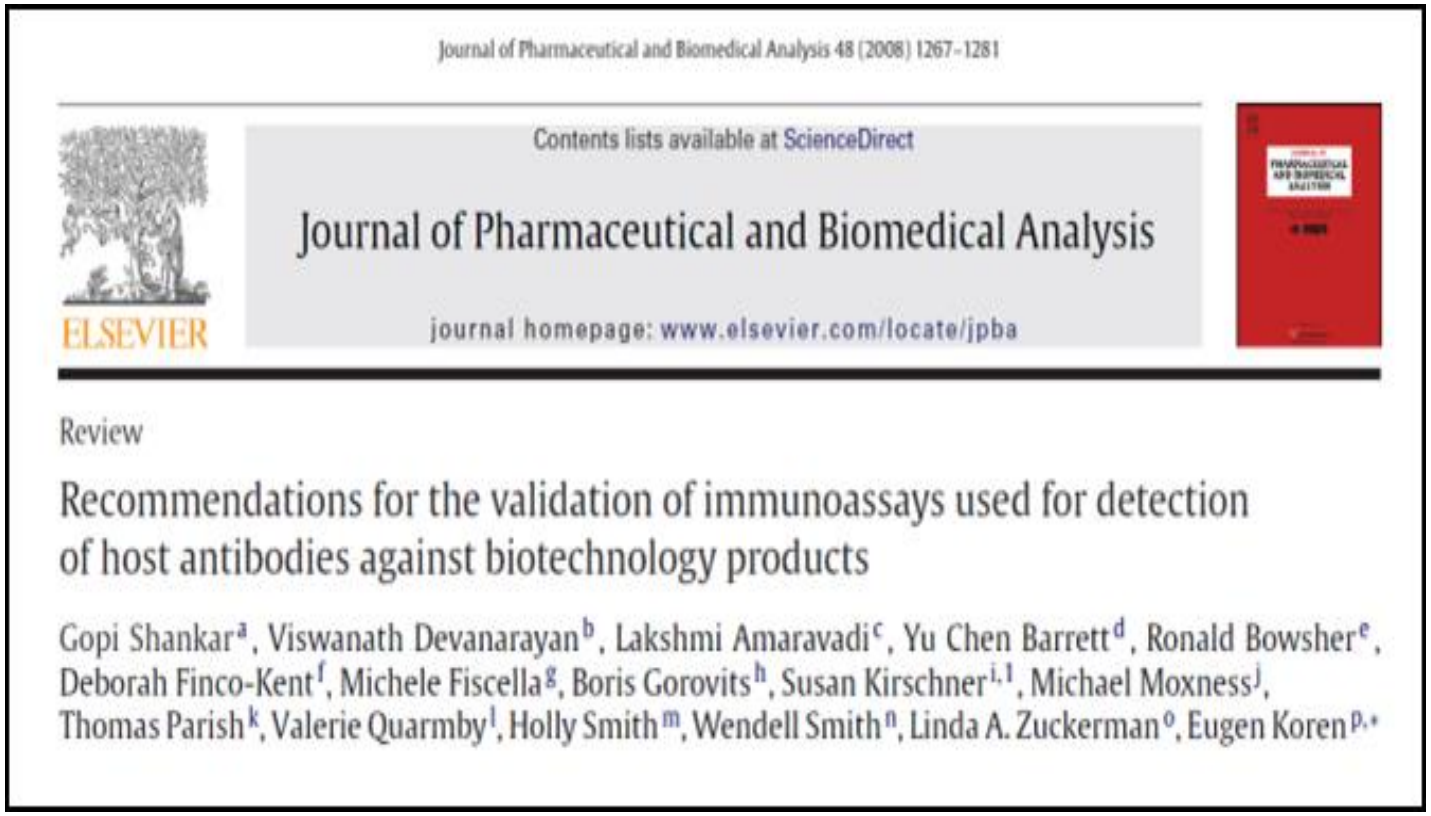
Recommendations such as those outlined in Shankar et al. 2008 have informed approaches to statistical analysis of immunogenicity data.

As the industry gains more experience, these approaches develop and change and revised recommendations such as those outlined in Devanarayan et al. 2017 suggest simplified approaches to these analyses.

There can be resistance to changes in approach.

LGC advocate a collaborative approach to statistical analysis and its application to immunogenicity data.

Shankar et al. 2008



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Review

Recommendations for the validation of immunoassays used for detection of host antibodies against biotechnology products

Gopi Shankar[a], Viswanath Devanarayan[b], Lakshmi Amaravadi[c], Yu Chen Barrett[d], Ronald Bowsher[e], Deborah Finco-Kent[f], Michele Fiscella[g], Boris Gorovits[h], Susan Kirschner[i,1], Michael Moxness[j], Thomas Parish[k], Valerie Quarmby[l], Holly Smith[m], Wendell Smith[n], Linda A. Zuckerman[o], Eugen Koren[p,*]

Devanarayan et al. 2017

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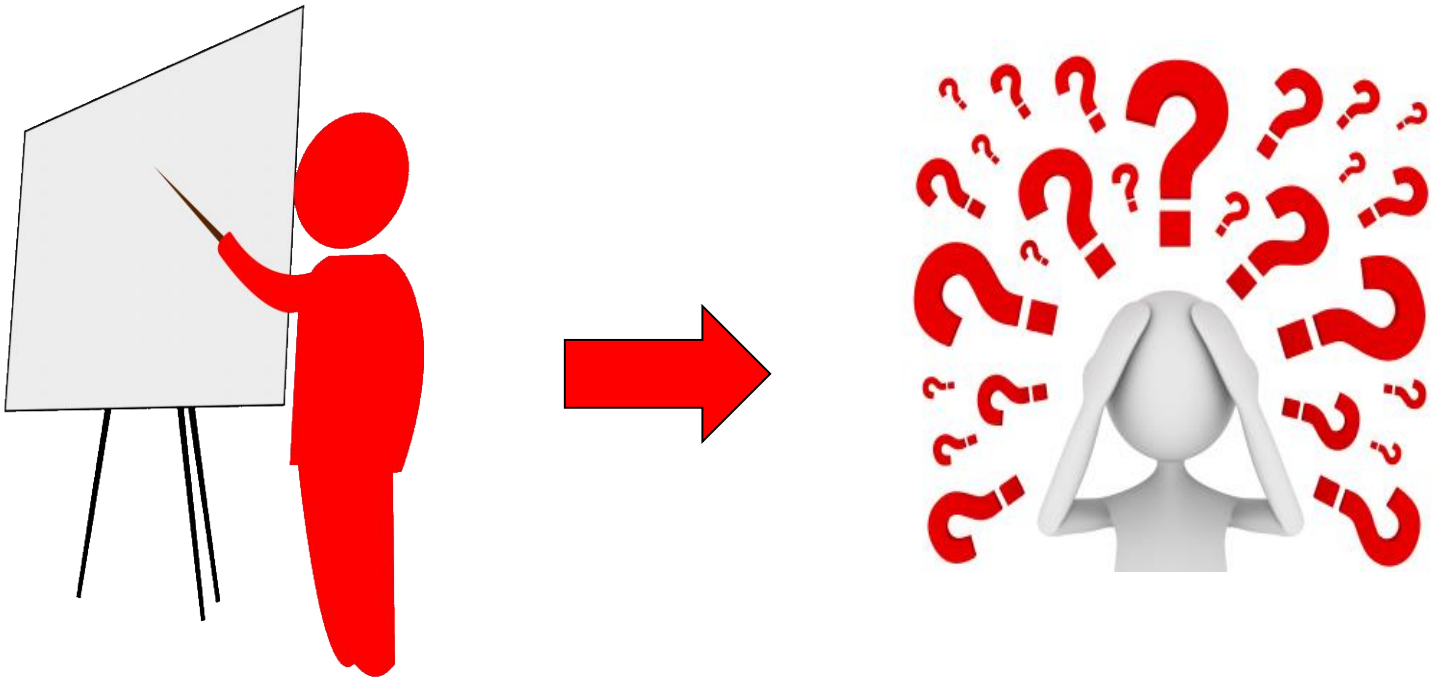
DOI: 10.1208/s12248-017-0107-3

Research Article

Recommendations for Systematic Statistical Computation of Immunogenicity Cut Points

Viswanath Devanarayan,[1] Wendell C. Smith,[2] Rocco L. Brunelle,[2] Mary E. Seger,[2,3] Kim Krug,[2,3] and Ronald R. Bowsher[2,3,4]

The initial reaction to a change in process …



Collaborative Process

Scientific decisions are supported by the additional information the statistician brings:

- underlying distribution of the data
- uncertainty in the estimates (of cut points etc.)

Statistician: implements the appropriate model to fit the data
Intermediary: an experienced scientist
Project scientist: understands science

Statistical decisions are aided by an understanding of the science – not just a numbers game

LGC CASE STUDIES

Based on real scenarios (mock data used to maintain client confidentiality)

Example #1
Initial experience with floating CCPs

Need for alternative approach

+ % inhibition for cut point individuals present and variable (ADDITION OF DRUG)

+ % inhibition for NCs present and variable (ADDITION OF DRUG)

variance in signal responses (IN ABSENCE OF DRUG)

	1	2	3	4	5	6	7	8	9	10	11	12
A	HPCa		IS6		IS14		IS1		IS9		IS17	
B	LPCa		IS7		IS15		IS2		IS10		IS18	
C	NCa		IS8		IS16		IS3		IS11		IS19	
D	IS1		IS9		IS17		IS4		IS12		IS20	
E	IS2		IS10		IS18		IS5		IS13		HPCa	
F	IS3		IS11		IS19		IS6		IS14		LPCa	
G	IS4		IS12		IS20		IS7		IS15		NeCa	
H	IS5		IS13		NCb		IS8		IS16		NCc	

Key

-DRUG **+DRUG**

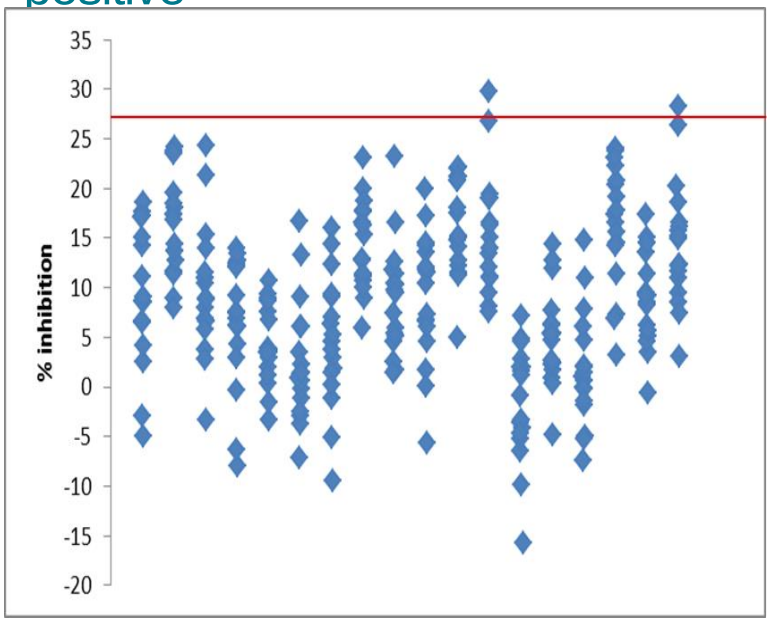
	1	2	3	4	5	6	7	8	9	10	11	12
A	8798	8550	450	452	914	973	83	103	85	77	81	75
B	145	142	162	149	525	544	70	70	81	95	74	62
C	108	112	140	139	152	161	72	84	79	89	72	66
D	391	349	140	145	543	528	69	68	73	73	67	72
E	155	154	255	246	210	156	67	85	92	89	76	70
F	222	216	160	149	127	122	66	72	81	86	72	62
G	1293	1348	191	186	153	144	72	81	88	66	69	64
H	1019	991	5517	5398	131	122	63	64	68	67	153	154

Signal responses

Need for alternative approach

Fixed CCP DID NOT WORK

- Individual samples do not have the same probability of being classified positive



← **Fixed CCP**

18 plates of data: 2 analysts x 3 runs (x3 plates)

- High False Positive rate >30%
- Inhibition is not constant across runs and plates
- NCs not consistently negative
- LPC (confirmatory) did not consistently confirm positive
- Normalised floating CCP allowed differentiation of positive and negative samples and controls with appropriate false positive rate.

Alternative Approach - normalisation

Inhibition ratio was calculated for each data point:

$$\frac{\text{(spiked/unspiked) sample or control}}{\text{(spiked/unspiked) NC}}$$

↓

Data sufficiently normal, no further transformation

↓

Box plot analysis assessed outliers (residuals)

↓

A random effects model was fitted to the data using PROC MIXED

↓

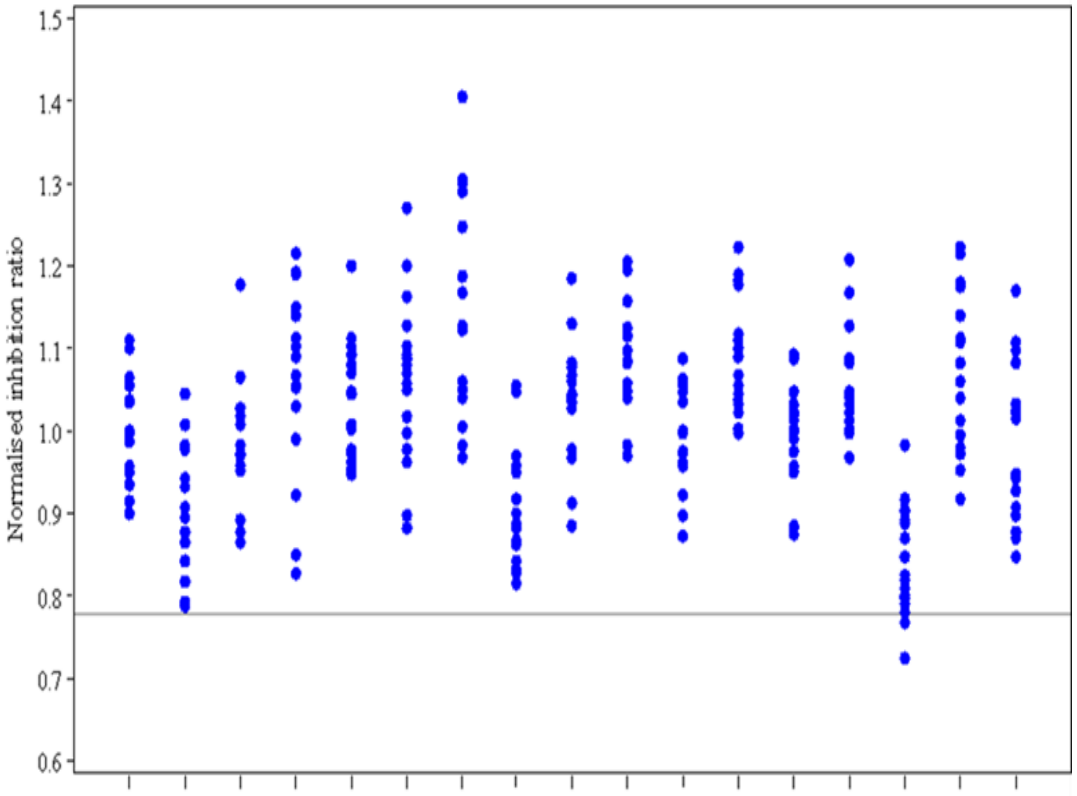
Standard deviation calculated, plots indicated a good fit of the model

↓

99% lower limit calculated which gave CCPF - gave False Positive Rate between 0.5 and 1%

Important factors

- Correlation between NC and sample means
- Variances must also be close
- We find that the run/plate variability is not always removed



Scientific and statistical input needed to decide when this approach may be necessary

Example #2
A new model for cut point calculation

Implementation

Our experience

- Advocacy by an experienced scientist for widespread acceptance and not just the statistician
- Drive to improve tolerance/sensitivity → lower variability→ questioning of new approaches (low CPFs distrusted)
- Acceptance of new approach to cut point setting is data driven

Implementing the new model:

- Statistician: provides understanding of method and writes SAS code for routine use
- Experienced Scientist: an experienced individual understands need for change and provides bridge between statistician and project scientists which leads to acceptance of new approaches

A new model for cut point calculation

Provides more information about:

- Outliers (biological versus analytical)
- Sources of variation
- Uses a statistical modelling approach easily implemented in SAS or other software
- Needs deeper knowledge of statistical methods

Comparison of Approach

- **Example: clinical validation, SCP calculation**
- **Devanarayan et al.: CPF = 1.09**
- **Shankar et al.: CPF = 1.16**

Outliers

	Not outlier	Outlier	**Total**
Not outlier	263		263
Analytical outlier	7	3	10
Biological outlier	9	20	29
Total	279	23	

Shankar et al.

False Positives

	Negative	Positive	**Total**
Negative	245		245
Analytical outlier	5	5	10
Biological outlier	21	8	29
Positive	12	6	18
Total	283	19	

Devanarayan et al.

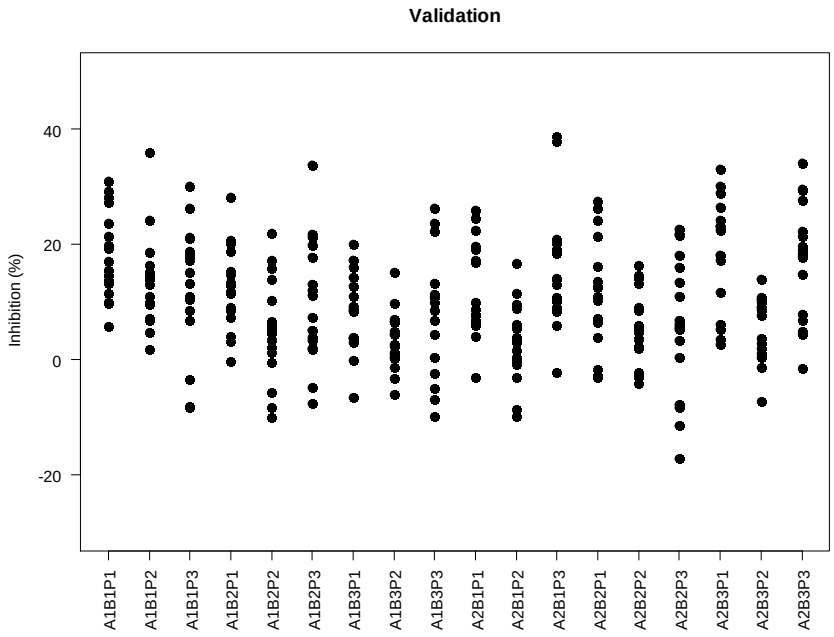
Shankar et al. vs. Devanarayan et al.

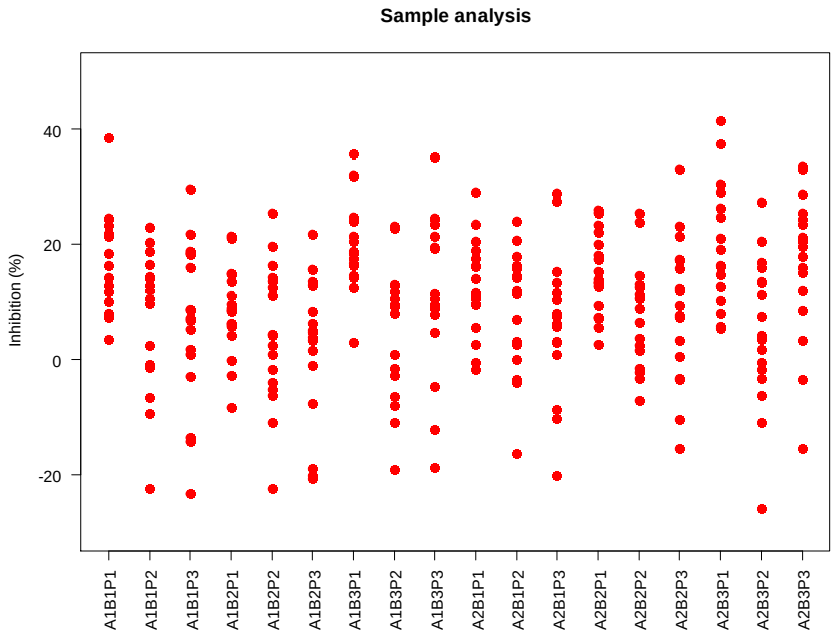
- Comparable False Positive Rates
- Shankar et al. approach – False Positive contribution from analytical and biological outliers
- Appeared to give the ‘right answer’ from scientist’s perspective
- Statistician’s model able to identify flaws in approach

Example #3
Do we need to change the cut point?

Validation vs. Sample analysis

- In-study data set will produce a cut point estimate which differs from the validation estimate
- Visually it looks as if there may have been a change in CP





Our view

- Only change a cut point it if there is a **demonstrable difference**
- This needs knowledge about uncertainty
- Scientist: assesses differences between validation and in-study populations for possible reasons why they could differ
- Statistician: compares CP estimates in terms of the uncertainty in the difference (i.e. is the change large enough?)

Uncertainty

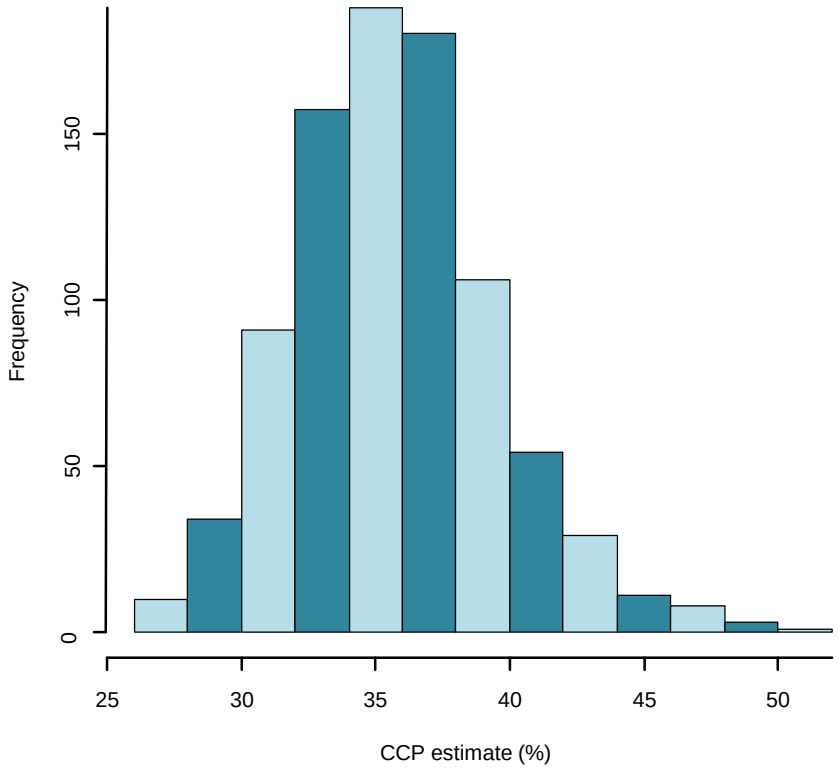
Uncertainty can be illustrated by Monte Carlo simulation

- Same population, many data sets
- Calculate CCP for each data set, look at distribution

CCP Estimates:

- Validation CCP = 33.8% inhibition
- In study CCP = 35.7% inhibition
- Is 1.9% significant? Or could random variation produce it?

Monte Carlo Simulation



Estimates have high degree of uncertainty therefore 1.9% loses significance

Only change the CP if it can be demonstrated that the means and/or variances (subject, run, plate) have changed – not by looking at the CP estimate – or if there are scientific grounds to do so

Conclusion

- Approaches in 2017 paper are being adopted
- Floating CCP can work well with good correlation between NC and sample means and similar variances
- Using old and new approaches for setting SCP may appear to give comparable answers – model can identify flaws
- Beware of comparing cut point calculations without understanding uncertainty
- Collaboration between Analytical Scientist and the Statistician is key to making the best decision

Simon Cowen,
Team Leader Statistics
(Science & Innovation)

Nicola Stacey and the
LGC Immunogenicity
Validation Teams (DDS)